

FORM-PTO-1390- (Rev. 10-96)		U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE		ATTORNEY'S DOCKET NUMBER	
<b>TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371</b>				001560-350	
				U.S. APPLICATION NO. (if known, see 37 C.F.R. 1.51) Unassigned <b>09/147955</b>	
INTERNATIONAL APPLICATION NO. PCT/JP98/03199		INTERNATIONAL FILING DATE 16 July 1998		PRIORITY DATE CLAIMED 25 July 1997	
TITLE OF INVENTION GENE CODING FOR A PROTEIN HAVING GLYCOSIDE TRANSFER ACTIVITY					
APPLICANT(S) FOR DO/EO/US Masako MIZUTANI, Yoshikazu TANAKA, Takaaki KUSUMI, Kazuki SAITO, Mami YAMAZAKI, and Gong ZHIZHONG					
Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:					
1.	<input checked="" type="checkbox"/>	This is a <b>FIRST</b> submission of items concerning a filing under 35 U.S.C. 371.			
2.	<input type="checkbox"/>	This is a <b>SECOND</b> or <b>SUBSEQUENT</b> submission of items concerning a filing under 35 U.S.C. 371.			
3.	<input checked="" type="checkbox"/>	This is an express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and the PCT Articles 22 and 39(1).			
4.	<input type="checkbox"/>	A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.			
5.	<input checked="" type="checkbox"/>	A copy of the International Application as filed (35 U.S.C. 371(c)(2))			
	a.	<input type="checkbox"/>	is transmitted herewith (required only if not transmitted by the International Bureau).		
	b.	<input checked="" type="checkbox"/>	has been transmitted by the International Bureau.		
	c.	<input type="checkbox"/>	is not required, as the application was filed in the United States Receiving Office (RO/US)		
6.	<input checked="" type="checkbox"/>	A translation of the International Application into English (35 U.S.C. 371(c)(2)).			
7.	<input checked="" type="checkbox"/>	Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))			
	a.	<input type="checkbox"/>	are transmitted herewith (required only if not transmitted by the International Bureau).		
	b.	<input type="checkbox"/>	have been transmitted by the International Bureau.		
	c.	<input type="checkbox"/>	have not been made; however, the time limit for making such amendments has NOT expired.		
	d.	<input checked="" type="checkbox"/>	have not been made and will not be made.		
8.	<input type="checkbox"/>	A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).			
9.	<input checked="" type="checkbox"/>	An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).			
10.	<input type="checkbox"/>	A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).			
<b>Items 11. to 16. below concern other document(s) or information included:</b>					
11.	<input checked="" type="checkbox"/>	An Information Disclosure Statement under 37 CFR 1.97 and 1.98.			
12.	<input checked="" type="checkbox"/>	An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.			
13.	<input checked="" type="checkbox"/>	A FIRST preliminary amendment.			
	<input type="checkbox"/>	A SECOND or SUBSEQUENT preliminary amendment.			
14.	<input type="checkbox"/>	A substitute specification.			
15.	<input type="checkbox"/>	A change of power of attorney and/or address letter.			
16.	<input checked="" type="checkbox"/>	Other items or information:			
Copy of International Search Report, and a copy of Notice Informing Applicant of the Communication of the International Application to the Designated Offices.					

U.S. APPLICATION NO. (If known, see 37 C.F.R. 1.50) <b>Unassigned</b>		INTERNATIONAL APPLICATION NO. <b>PCT/JP 98/03199</b>		ATTORNEY'S DOCKET NUMBER <b>001560-350</b>	
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17. <input checked="" type="checkbox"/> The following fees are submitted:				<b>CALCULATIONS</b>		PTO USE ONLY	
<b>Basic National Fee (37 CFR 1.492(a)(1)-(5)):</b>  Search Report has been prepared by the EPO or JPO ..... \$840.00  International preliminary examination fee paid to USPTO (37 CFR 1.482) ..... \$670.00 No international preliminary examination fee paid to USPTO (37 CFR 1.482) but international search fee paid to USPTO (37 CFR 1.445(a)(2)) ..... \$760.00  Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO ..... \$970.00  International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(2)-(4) ..... \$96.00							
<b>ENTER APPROPRIATE BASIC FEE AMOUNT =</b>							
Surcharge of \$130.00 for furnishing the oath or declaration later than months from the earliest claimed priority date (37 CFR 1.492(e)). <span style="float: right;"><input type="checkbox"/> 20 <input type="checkbox"/> 30</span>							
Claims	Number Filed	Number Extra	Rate				
Total Claims	19 -20 =	0	X\$18.00	\$	0.00		
Independent Claims	1 -3 =	0	X\$78.00	\$	0.00		
Multiple dependent claim(s) (if applicable)			+ \$260.00	\$	0.00		
<b>TOTAL OF ABOVE CALCULATIONS =</b>				\$	840.00		
Reduction for 1/2 for filing by small entity, if applicable. Verified Small Entity statement must also be filed. (Note 37 CFR 1.9, 1.27, 1.28).				\$	0.00		
<b>SUBTOTAL =</b>				\$	840.00		
Processing fee of \$130.00 for furnishing the English translation later than months from the earliest claimed priority date (37 CFR 1.492(f)). <span style="float: right;"><input type="checkbox"/> 20 <input type="checkbox"/> 30</span>				\$	0.00		
<b>TOTAL NATIONAL FEE =</b>				\$	840.00		
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). \$40.00 per property +				\$	40.00		
<b>TOTAL FEES ENCLOSED =</b>				\$	880.00		
				Amount to be:			
				refunded		\$	
				charged		\$	

a. ☒ A check in the amount of \$ 880.00 to cover the above fees is enclosed.

b. ☐ Please charge my Deposit Account No. 02-4800 in the amount of \$ \_\_\_\_\_ to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. 02-4800. A duplicate copy of this sheet is enclosed.

**NOTE:** Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

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Donna M. Meuth  
 NAME

36,607  
 REGISTRATION NUMBER

09/147955  
510 Rec'd PCT/PTO 24 MAR 1999

Patent  
Attorney's Docket No. 001560-350

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

In re Patent Application of )  
)  
Masako MIZUTANI et al ) Group Art Unit: Unassigned  
)  
Application No.: Unassigned ) Examiner: Unassigned  
Corresponding to PCT/JP 98/03199 )  
)  
Filed: March 24, 1999 )  
)  
For: GENE CODING FOR A PROTEIN )  
HAVING GLYCOSIDE TRANSFER )  
ACTIVITY )

**PRELIMINARY AMENDMENT**

Assistant Commissioner for Patents  
Washington, D.C. 20231

Sir:

Prior to examination on the merits, please amend the above identified application as follows:

**IN THE SPECIFICATION:**

In compliance with 37 C.F.R. § 1.823(a), please substitute the attached copy of the "Sequence Listing" for the current "Sequence Listing" at pages 22-39 of the above-identified application.

**IN THE CLAIMS:**

Please amend claims 6, 8 and 10 as follows:

In claim 6, lines 1 and 2, please delete "any one of claims 1 through 5" and insert therefore --claim 1--.

In claim 8, lines 1 and 2, please delete "any one of claims 1 through 5" and insert therefore --claim 1--.

In claim 10, line 2, please delete "any one of claims 1 through 5" and insert therefore --claim 1--.

Please insert the following new claims 12-19 as follows:

--12. A protein encoded by a gene as set forth in claim 2.

13. A protein encoded by a gene as set forth in claim 3.

14. A protein encoded by a gene as set forth in claim 4.

15. A protein encoded by a gene as set forth in claim 5.

16. A plant into which is introduced a gene as set forth in claim 2, or its progeny or tissue having identical properties.

17. A plant into which is introduced a gene as set forth in claim 3, or its progeny or tissue having identical properties.

18. A plant into which is introduced a gene as set forth in claim 4, or its progeny or tissue having identical properties.

19. A plant into which is introduced a gene as set forth in claim 5, or its progeny or tissue having identical properties.--

**REMARKS**

Entry of the foregoing and examination of the above-identified application is respectfully requested.

The paper copy of the Sequence Listing for the subject application, is by this amendment, substituted for the current Sequence Listing at pages 22-39 and before the claims of the above-identified application. Please renumber the pages accordingly.

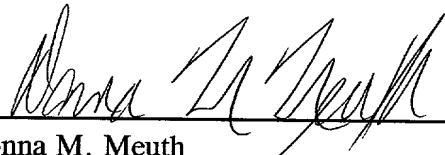
Claims 6, 8 and 10 have been amended to eliminate the multiple dependency of the claims and to place them in better form in accordance with U.S. practice. New claims 12-19 have been added directed to preferred embodiments. Support for these claims may be found at the very least in original claims 8 and 10.

Early and favorable action in the form of Notice of Allowance is respectfully requested.

In the event that there are any questions relating to this amendment or the application in general, it would be appreciated if the Examiner would contact the undersigned attorney by telephone so that prosecution would be expedited.

Respectfully submitted,

BURNS, DOANE, SWECKER & MATHIS, L.L.P.

By:   
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Date: March 24, 1999

GENE CODING FOR A PROTEIN HAVING GLYCOSIDE TRANSFER  
ACTIVITY

5 Technical Field

The present invention relates to a gene coding for a protein having activity that transfers a glycoside to the 5 position of a flavonoid, and a process utilizing that gene.

10

Background Art

The flower industry strives to develop various new varieties. Changing the color of a flower is one way of effectively breeding a new variety. A wide range of colors have been successfully produced for nearly all commercial varieties using classical breeding methods. With these methods, however, since there are restrictions on the gene pool for each species, it is rare for a single species to have a broad range of colored varieties.

20 Flower colors are based on two types of pigments, namely flavonoids and carotinoids. Flavonoids contribute to color tones ranging from yellow to red and blue, while carotinoids contribute to color tones of orange or yellow. Flavonoid molecules that primarily contribute to flower color are anthocyanins which are glycosides of cyanidin, delphinidin, petunidin, peonidin, malvidin and pelargonidin, and different anthocyanins cause remarkable changes in flower color. Moreover, flower color is also affected by auxiliary coloring by colorless flavonoids, metal complex formation, glucosylation, acylation, methylation and vacuolar pH (Forkmann, Plant Breeding, 106, 1, 1991).

25 The biosynthesis route of anthocyanins, which begins with phenylalanine, has been well understood (e.g., Plant Cell, 7, 1071-1083, 1995), and nearly all genes involved in the biosynthesis have been cloned. For example, among those genes thought to be involved in biosynthesis of

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malonylshisonin (3-0-(6-0-(p-cumaloyl)- $\beta$ -D-glucosyl)-5-0-(6-0-malonyl- $\beta$ -D-glucosyl)-cyanidin), which is an anthocyanin of Perilla, those genes for which homologues have not yet been reported are only the flavonoid-3'-hydroxylase, UDP-glucose:anthocyanin (flavonoid) 5-0-glucosyl transferase (abbreviated as 5GT) and malonyl group transferase genes.

Among these, flavonoid-3'-hydroxylase is known to belong to the cytochrome P450 gene family (Plant Cell, 7, 1071-1083, 1995), and cytochrome P450 genes are surmised to demonstrate structural homology.

The hydroxyl group at the 3 position of flavonoid molecules is typically modified by glucose, and generally glucosylation and other modifications by glycoside are considered to increase the stability and solubility of anthocyanins (The Flavonoids, Chapman & Hall, 1994).

Genes coding for the UDP-glucose:anthocyanidin or flavonoid-3-glucosyl transferase (abbreviated as 3GT) that catalyze this reaction are obtained from numerous plants such as corn, barley, snapdragons and gentians, and their amino acid sequences mutually demonstrate significant homology. For example, the homology between the 3GT amino acid sequences of monocotyledonous corn and dicotyledonous gentian is 32%, that between the 3GT amino acid sequences of monocotyledonous corn and monocotyledonous barley is 73%, and that between the 3GT amino acid sequences of dicotyledonous gentian and dicotyledonous eggplant is 46%.

In addition, the gene coding for UDP-ramnose:anthocyanidin 3-glucosidoramnosyl transferase (3RT) of petunias has also been cloned.

However, even though the hydroxyl group at the 5 position of the flavonoids of numerous plants is glucosylated, a gene for the enzyme (5GT) that catalyzes this reaction has yet to be obtained.

In addition, although there are examples of measuring the reaction by which glycoside is transferred to the 5



position of petunia and stock anthocyanins (Planta, 160, 341-347, 1984, Planta, 168, 586-591, 1986), these reports only describe the investigation of enzymological properties using crude extracts or partially purified products of flower petals, and there are no examples of this enzyme being purified to its pure form. In addition, since glycosyltransferases are typically biochemically unstable, enzyme purification is difficult.

Although there are hardly any cases in which color tone is changed by addition of glycoside to a flavonoid molecule, since aromatic acyl groups that have a significant effect on color tone are linked to a glucose molecule or rhamnose molecule within an anthocyanin, regulation of the glycoside transfer reaction is important in terms of controlling anthocyanin biosynthesis, and ultimately in controlling flower color. Furthermore, as an example of changing flower color by regulating the expression of glycosyltransferase gene, the reaction by petunia 3RT has been controlled in transformed petunia to modify flower color.

Plant species, which can be transformed with a foreign gene, include, for example, roses, chrysanthemums, carnations, daisies, petunias, torenia, bellflowers, calanchoes, tulips and gladiolas.

#### Disclosure of the Invention

The inventors of the present invention therefore sought to obtain a gene that codes for a protein having activity that transfers a glycoside to the 5 position of a flavonoid, thereby leading to completion of the present invention.

For example, the 5 position hydroxyl group of the anthocyanins of chrysanthemums and some of the anthocyanins of roses and carnations are not glucosylated. The anthocyanin structure can be changed by introducing the 5GT gene obtained by the present invention into these plants.

In addition, although it is possible to change flower color and stabilize flavonoids by acylating flavonoids using the acyl group transferase gene described in International Publication No. WO96/25500, since the acyl group does not bond directly with the flavonoid, but rather bonds by way of a sugar, simply introducing an acyl group transferase gene alone is not sufficient for changing flower color and may even cause the flavonoid not to become stable.

However, by introducing the 5GT gene in combination with an acyl group transferase gene, sugar is bounded to the 5 position of the flavonoid thereby further allowing the flavonoid to be acylated. This can be expected to change the anthocyanin structure and cause the flower color to become bluish.

In addition, if expression of 5GT gene of a plant in which the 5 position of anthocyanin is glucosylated is suppressed with the antisense method or co-suppression method and so forth, transfer of glucose residue to 5 position can be inhibited. So that, flower color can be changed. For example, suppressing 5GT activity in gentian or bellflower can be expected to cause flower color to become reddish.

The inventors of the present invention isolated cDNA of 5GT from Perilla, torenia, verbena and petunia plants using gene recombination technology, and determined the nucleotide sequence of the structural gene. Namely, the inventors of the present invention provide a DNA sequence that codes for 5GT present in the tissue that expresses anthocyanins in these plants. Moreover, since this enzyme transfers glycoside to the 5 position of anthocyanin pigment, it can be used to change flower color and increase anthocyanin stability.

#### Embodiment for Carrying Out the Invention

The method of differential displacement, for example, can be used to obtain DNA that codes for the enzyme of the

present invention. In Perilla (Perilla frutescens), for example, there are varieties that accumulate anthocyanins (e.g., red forma) and those that do not (e.g., green forma). By cloning DNA present in varieties that  
5 accumulate anthocyanins but not present in varieties that do not, it is possible to obtain the DNA that codes for the enzyme of the present invention.

More specifically, RNA is extracted from the leaves of red forma and green forma, and cDNA is synthesized in  
10 accordance with standard methods. This is then separated by electrophoresis to isolate cDNA present in the cDNA library of red forma but not present in the cDNA library of green forma. Next, the red forma cDNA library is screened using the resulting cDNA as a probe to obtain the  
15 cDNA that codes for the enzyme of the present invention.

Once cDNA that codes for the enzyme of the present invention is obtained in the manner described above, this cDNA or its fragment is used as a probe to screening the cDNA libraries of other plants. As a result, the DNA that  
20 codes for the enzyme of the present invention can be obtained from those plants.

As an example of the screening, in the present invention, the DNA coding for the enzyme of the present invention is cloned from Perilla by the differential  
25 display method (Example 1). Next, DNA that codes for the enzyme of the present invention is obtained from verbena by screening of cDNAs from verbena (Verbena hybrida) using the cloned DNA of Example 1 as a probe (Example 2). Moreover, DNA coding for the enzyme of the present  
30 invention is obtained from torenia in the same manner (Example 3).

Then, it was confirmed that the proteins encoded in these DNAs have the enzymatic activity of the present invention.

35 Moreover, the DNA coding for the enzyme of the present invention was obtained from petunia (Example 4).

Examples of the DNAs of the present invention include

that which codes for the amino acid sequence described in any one of SEQ ID NOs: 7 through 10 or 12. However, proteins having an amino acid sequence modified by addition and/or deletion of one or more amino acids and/or substitutions by one or more other amino acids are also known to maintain enzymatic activity similar to the original protein. Thus, genes coding for a protein that has an amino acid sequence modified by addition and/or deletions of one or more amino acids and/or substitutions by one or more other amino acids relative to the amino acid sequence described in any one of SEQ ID NOs: 7 through 10 or 12, and still maintains activity of transferring a glycoside to the 5 position of a flavonoid, also belong to the present invention.

The present invention also relates to a gene coding for a protein which gene hybridizes to a nucleotide sequence described in any one of SEQ ID NOs: 1 through 4 or 6, or to a nucleotide sequence that codes for an amino acid sequence described therein or to their portions, for example a portion coding for at least six amino acids of a consensus region, under conditions of 2 to 5 x SSC, and for example, 5 x SSC, and 50°C, and that has activity of transferring a glycoside to the 5 position of a flavonoid. Furthermore, the optimum hybridization temperature varies according to the nucleotide sequence and its length, and it is preferable that the hybridization temperature be lower the shorter the nucleotide sequence. For example, a temperature of 50°C or lower is preferable in the case of a nucleotide sequence (18 bases) coding for six amino acids.

Although examples of genes selected by hybridization in this manner include those which are naturally-occurring such as those derived from plants, examples of which include a gene derived from verbena and torenia, they may also be those derived from other plants, examples of which include petunias, roses, carnations and hyacinths. In addition, genes selected by hybridization may also be cDNA or genomic DNA.

Moreover, the present invention also relates to a gene coding for a protein having an amino acid sequence having homology of 30% or more, preferably 50% or more, for example 60% or 70% or more, and in some cases, 90% or more relative to an amino acid sequence of any of SEQ ID NOs: 7 through 10 or 12, and having activity that transfers a glycoside to the 5 position of a flavonoid. Namely, as indicated in Examples, DNA coding for the enzyme of the present invention demonstrates homology of 20 to 30% in comparison with other glycosyltransferase genes. Thus, the present invention includes genes coding for a protein that having homology of 30% or more with an amino acid sequence described in any one of SEQ ID NOs: 7 through 10 or 12, and has glycosyltransferase activity.

In addition, as is clear from a comparison of the results of Examples 1 through 4, the amino acid sequence of the enzyme of the present invention varies according to the species, with interspecies homology being 50% or more (see Examples 3 and 4), and for example 60 to 70% (see Example 2), while the homology of the amino acid sequences of the enzymes derived from the same species is 90% or more (see Example 1). Thus, genes coding for a protein that has an amino acid sequence having homology of 50% or more, for example 60-70% or more, and in some cases, 90% or more, relative to an amino acid sequence described in any one of SEQ ID NOs: 7 through 10 or 12, and maintains the glycosyltransferase activity of the present invention are included in the present invention.

As is described in detail in Examples, DNA having a native nucleotide sequence is obtained by, for example, screening of a cDNA library.

In addition, DNA coding for an enzyme having a modified amino acid sequence can be synthesized using ordinary site-specific mutagenesis and PCR based on the nucleotide sequence of a native DNA. For example, a DNA fragment containing a site at which a modification is desired to be introduced is obtained by restriction enzyme

digestion of cDNA or genomic DNA obtained as described above. By using this as a template, site-specific mutagenesis or PCR is performed using a primer containing the desired mutation to obtain a DNA fragment containing the desired modification. This is then ligated to DNA coding for another portion of the target enzyme.

Alternatively, in order to obtain DNA coding for an enzyme having a shortened amino acid sequence, for example, DNA coding for an amino acid sequence that is longer than the target amino acid sequence, for example that coding for the entire amino acid sequence, is digested by a desired restriction enzyme, and in the case the resulting DNA fragment does not code for the entire target amino acid sequence, the deficient portion should be supplemented by ligating synthetic DNA.

In addition, by expressing this clone using a gene expression system in E. coli or yeast and measuring enzyme activity, the resulting gene can be confirmed to code for glycosyltransferase, and by clarifying the translation region of glycosyltransferase gene that transfers glycoside to the 5 position of a flavonoid, a gene is obtained that codes for the glycosyltransferase claimed in the present invention. Moreover, by expressing said gene, the target transferase protein that transfers a glycoside to the 5 position of a flavonoid can be obtained.

Alternatively, the protein can be obtained by using antibody to an amino acid sequence described in any one of SEQ ID NOs: 7 through 10 or 12.

Thus, the present invention also relates to a recombinant vector containing the above-mentioned DNA, and more particularly, to an expression vector and a host transformed with the vector. Both prokaryotes and eukaryotes can be used for the host. Examples of prokaryotes that can be routinely used for the host include bacteria, for example, the genus Escherichia such as Escherichia coli, and the genus Bacillus such as Bacillus subtilis.

Examples of eukaryotes that can be used include lower eukaryotes such as eucaryotic microorganisms including fungi such as yeast or mold. Examples of yeast includes the genus Saccharomyces such as Saccharomyces cerevisiae,  
5 while examples of molds include the genus Aspergillus such as Aspergillus oryzae and Aspergillus niger, as well as the genus Penicillium. Moreover, animal or plant cells can also be used, examples of animal cells including mouse, hamster, monkey and human cell systems. Moreover,  
10 insect cells such as silkworm cells or adult silkworms themselves can be used as hosts.

The expression vectors of the present invention contain an expression control region, such as a promoter, terminator or an origin of replication, depending on the  
15 type of host in which they are to be introduced. Examples of promoters of bacterial expression vectors include conventionally used promoters such as trc promoter, tac promoter and lac promoter, while examples of yeast promoters include glyceroaldehyde triphosphate  
20 dehydrogenase promoter and PH05 promoter. Examples of mold promoters include amylase and trpC. In addition, examples of promoters for animal cell hosts include viral promoters such as SV40 early promoter and SV40 late promoter.

25 Preparation of expression vector can be performed in accordance with standard methods using restriction enzyme, ligase and so forth. In addition, transformation of a host by an expression vector can also be performed in accordance with standard methods.

30 In the process for producing the above-mentioned protein, a host transformed with the expression vector is cultured, cultivated or bred, the target protein can be recovered and purified from the resulting culture in accordance with standard methods, examples of which  
35 include filtration, centrifugation, cell homogenation, gel filtration chromatography and ion exchange chromatography.

Furthermore, although the present specification

describes transferases derived from Perilla, verbena, torenia and petunia wherein the transferases that transfer glycoside to the 5 position of a flavonoid (which may be simply referred to as "glycosyltransferase" in the present invention), a gene that codes for said enzyme can be cloned, by entirely or partially altering the purification method of said enzyme so as to purify a glycosyltransferase of another plant, and determining the amino acid sequence of said enzyme. Moreover, by using cDNA of the glycosyltransferase derived from Perilla of the present invention as a probe, cDNA of a different glycosyltransferase was able to be obtained from Perilla, and cDNA of a different glycosyltransferase was able to be obtained from a different plant. Thus, other glycosyltransferase genes can be obtained by using a portion or the entirety of a glycosyltransferase gene.

In addition, as indicated in the present specification, by purifying glycosyltransferase from Perilla, verbena, torenia and petunia to obtain antibody to said enzyme in accordance with standard methods, cDNA or chromosomal DNA produces protein which reacts with that antibody that can be cloned. Thus, the present invention is not limited to only genes of glycosyltransferases derived from Perilla, verbena, torenia and petunia, but also relates to glycosyltransferase in the broad sense.

Moreover, the present invention also relates to a plant, its progeny or their tissue for which color has been adjusted by introduction of glycosyltransferase gene, and their form may be that of cut flowers as well.

In addition, UDP-glucose is an example of a glycoside donor in the glycoside transfer reaction of glycoside that include anthocyanin in the present specification.

#### Examples

The following provides a detailed explanation of the present invention based on Examples. Unless specified otherwise, the experimental procedure was performed in



accordance with the methods described in Molecular Cloning (Cold Spring Harbor, 1989), New Biochemistry Experimental Manual (Kagaku Dojin, 1996) and International Patent Laid-Open Publication No. WO96/25500.

5        Example 1 Cloning of a Gene Specifically Expressed in  
         Red Forma

(1) Differential Display

Perilla (Perilla frutescens) includes varieties that accumulate anthocyanins in their leaves (for example, red  
10 forma (Sakata-no-tane)), and varieties that do not accumulate anthocyanins (for example, blue forma (Sakata-no-tane)). The structure of the major anthocyanin is reported to be malonylshisonin (3-0-(6-0-(p-cumaloyl)- $\beta$ -D-glucosyl)-5-0-(6-0-malonyl- $\beta$ -D-glucosyl)-cyanidin) (Agri.  
15 Biol. Chem., 53:197-198, 1989).

Differential display is a method reported in Science, 257, 967-971 (1992), and is used, for example, to obtain genes that are expressed tissue-specifically.

Total RNA was extracted from the leaves of the above-mentioned two types of Perilla by the hot phenol method  
20 (Plant Molecular Biology Manual, Kluwer Academic Publishers, 1994, pp. D5/1-13). Poly A + RNA was purified from the resulting total RNA using an mRNA separator kit (Clontech). 0.9  $\mu$ g of poly A + RNA were reverse-  
25 transcribed in 33  $\mu$ l of reaction mixture using oligo-dT primer added an anchor (GenHunter, H-T11G, H-T11A and H-T11C) to obtain single strand cDNA. Using this cDNA as a template, PCR was performed using the same oligo-dT primer added an anchor and synthetic primers (GenHunter, H-AP1  
30 through 8) as primers.

The volume of the PCR reaction mixture was 20  $\mu$ l, and it contained 2  $\mu$ l of cDNA solution, 0.2  $\mu$ M of any one of H-T11G, H-T11A or H-T11C primer, 0.2  $\mu$ M of any primer from H-AP1 through H-AP8, 0.12  $\mu$ M dNTP, 5 or 10  $\mu$ Ci of  
35 [<sup>32</sup>P]dCTP, 10 mM Tris-HCl (pH 9.0), 50 mM KCl, 0.01% Triton X-100, 1.25 mM MgCl<sub>2</sub> and 1 unit of Taq polymerase. The reaction conditions comprised holding the temperature at

72°C for 20 seconds followed by repeating the reaction for 40 cycles with one cycle comprising raising the temperature to 94°C for 30 seconds, lowering to 40°C for 2 minutes and raising to 72°C for 30 seconds, and then  
5 holding the temperature at 72°C for 5 minutes.

The DNA fragments amplified in this manner were separated by the same polyacrylamide gel electrophoresis as used for DNA Sequencing. After drying the gel, the gel was exposed to X-ray film. Among the resulting  
10 approximately 2,600 bands, there were 36 bands observed only in the red forma as a result of comparing the two varieties. They were cut out of the dried gel and eluted into 100 µl of water. The eluted DNA was precipitated with ethanol and dissolved in 20 µl of water. Using a  
15 half amount of each DNA as a template, the PCR reaction was performed as described above, and amplified fragments were obtained for 33 of DNA fragments. Library screening and northern analysis were then performed using these DNA fragments.

## 20 (2) Northern Analysis

Northern analysis was performed according to the method described below using the above 33 types of DNA probes. After separating poly A + RNA derived from red forma and green forma with formamide gel containing 1.2%  
25 agarose, the poly A + RNA was transferred to a Nylon membrane. This membrane was hybridized with the above-mentioned DNA probes labeled with [<sup>32</sup>P] for overnight at 65°C in the presence of 5XSSPE, 5X Denhalt's solution, 0.5% SDS and 20 µg/ml of denatured salmon sperm DNA. The  
30 hybridized membrane was washed at 65°C in 1XSSPE and 0.1% SDS solution and subjected to autoradiography. As a result, only five probes were specifically expressed in red forma. These clones are predicted to be genes involved in the biosynthesis of anthocyanins.

## 35 (3) Screening of cDNA Library

A cDNA library with λgt10 as a vector was prepared using the poly A + RNA obtained from the leaves of red

forma and the Complete Rapid Cloning System  $\lambda$ gt10 (Amersham). This cDNA library was screened with the five DNA fragments described above to obtain cDNA corresponding to each fragment. Among these, a clone named 3R5 was  
5 obtained using a DNA fragment obtained by H-T11A and H-AP3 primers, and this clone demonstrated homology of approximately 26% at the amino acid level with previously reported corn flavonoid-3-O-glucosyl transferase.

In addition, clones designated as 3R4 and 3R6 were  
10 obtained by library screening using the same probes, and these demonstrated an extremely high level of homology with 3R5. The complete nucleotide sequences and deduced amino acid sequences of 3R4 and 3R6 are shown in SEQ ID NO: 1 and SEQ ID NO: 2 of the Sequence Listing,  
15 respectively. In addition, the deduced amino acid sequences of the proteins encoded by 3R4 and 3R6 demonstrated homology of 92%.

A clone designated as 8R6 was obtained using a DNA fragment obtained by H-T11G and H-AP8 primers, and this  
20 clone did not demonstrate significant homology with any sequences reported so far. This sequence is shown in SEQ ID NO: 5 of the Sequence Listing. Although there is a strong possibility that 8R6 is a gene involved in the biosynthesis of anthocyanins, since its structure lacks  
25 homology with genes reported so far, it is predicted to be a new gene involved in anthocyanin biosynthesis.

In consideration of the anthocyanin structure in Perilla (the previously mentioned malonylshisonin), it is predicted that this gene is a malonyl transferase. In  
30 order to verify this, this gene should be expressed in yeast and *E. coli* followed by reacting with anthocyanin and malonyl-CoA as substrates. Such an experiment can be carried out using, for example, the method described in International Publication No. WO96/25500. Malonyl  
35 transferase gene is useful in terms of artificially altering anthocyanin structure.

(4) Expression of 3R4 cDNA in Yeast

An approximately 1.5 kb DNA fragment obtained by blunting the BstXI cleaved site of p3R4 using T4 DNA polymerase (Takara Shuzo) and then cutting out at the BamHI cleavage site in the adapter, and an approximately 8  
5 kb DNA fragment obtained by blunting the EcoRI cleaved end of pYE22m and then digesting with BamHI were ligated to obtain a plasmid that was designated as pY3R4.

Furthermore, E. coli strain JM109 having pYE22m was named Escherichia coli SBM335, and deposited at the  
10 National Institute of Bioscience and Human-Technology Agency of Industrial Science and Technology as FERM BP-5435. In pY3R4, cDNA coding for glycosyltransferase has been ligated downstream of the promoter for  
glyceroaldehyde triphosphate dehydrogenase lone of the  
15 constitutive yeast promoter, and transcription is controlled by this promoter.

Using pY3R4, yeast Saccharomyces cerevisiae G1315 (Ashikari, et al., Appl. Microbiol. Biotechnol., 30, 515-520, 1989) was transformed according to the method of Ito, et al. (Ito, et al., J. Bacteriol., 153, 163-168, 1983).  
20 The transformed yeast was selected according to recovery of tryptophan synthesis ability. The resulting transformed strain was cultured for 24 hours at 30°C with shaking in 10 ml of Burkholder's medium (Burkholder, Amer. J. Bot., 30, 206-210) containing 1% casamino acids.  
25

In order to conduct a control experiment, yeast that spontaneously recovered tryptophan synthesis ability was also cultured in the same manner. After collecting the yeast, the cells were suspended in suspension buffer (100  
30 mM phosphate buffer (pH 8.5), 0.1% (v/v) 2-mercaptoethanol, 10  $\mu$ M APMSF and 100  $\mu$ M UDP-glucose) followed by the addition of glass beads (Glass Beads, 425-600 microns Acid-Wash, Sigma) and vigorous shaking to crush the cells. The crushed cells were then centrifuged  
35 for 20 minutes at 15,000 rpm and the supernatant was used as a crude enzyme solution for the measurement of enzyme activity described below.

(5) Measurement of Enzymatic Activity

After allowing 50  $\mu$ l of reaction mixture containing 20  $\mu$ l of crude enzyme solution (100 mM phosphate buffer (pH 8.5), 670  $\mu$ M cyanidin-3-glucoside, 1 mM UDP-glucose) for 10 minutes at 30 °C, 50  $\mu$ l of 50% acetonitrile solution containing 0.1% TFA was added to stop the reaction. Supernatant obtained by centrifuging for 5 minutes at 15,000 rpm was passed through a Samprep LCR4(T)-LC filter (Millipore) so as to remove impurities. This was then analyzed by high-performance liquid chromatography (HPLC). Analysis was performed using a reverse phase column (Asahipak ODP-50, 4.6 mm diameter x 250 mm, Showa Denko), the mobile phase consisted of 0.5% TFA/H<sub>2</sub>O for solution A and 0.5% TFA 50% CH<sub>3</sub>CN for solution B. The flow rate was 0.6 ml/min. and the fractions were eluted at a gradient of B20%  $\rightarrow$  B100% (20 min) followed by holding at B100% for 5 minutes.

20  $\mu$ l of reaction mixture was used for analysis. A520 nm, AUFS 0.5 (Shimadzu SPD-10A) and a photodiode array detector (Shimadzu SPD-M6A) at an absorbance of 600-250 nm were used for detection. In the case of reaction of yeast crude enzyme solution that expressed pY3R4, in addition to the substrate cyanidin-3-glucoside (retention time: 17 minutes), a new peak was observed at retention time of 14.5 minutes. Since it was not observed in the case of reaction of yeast crude enzyme solution of the control experiment, this new peak was considered to be generated due to the activity of protein originated from pY3R4. As a result of co-chromatography with cyanidin-3,5-diglucoside, the retention time of this peak coincided with that of cyanidin-3,5-diglucoside, and their absorption spectra were also identical to each other. Based on these observations, 3R4 cDNA of Perilla was found to code for 5GT.

Example 2 Cloning of 5GT Gene of Verbena hybrida

(1) Preparation of cDNA Library

Petals were collected from Verbena variety Hanatemari

64420"55624T60

violet (Suntory) and ground by a mortar and pestle in liquid nitrogen. RNA was extracted from the ground tissues according to a method using guanidine thiocyanate/cesium chloride, and poly A + RNA was obtained by the method recommended by the manufacturer using Oligotex (Takara Shuzo). The method using guanidine thiocyanate/cesium chloride was carried out in accordance with the method described in detail in Methods in Molecular Biology, Vol. 2 (Humana Press Inc., 1984) by R. McGookin and Robert J. Slater, et al.

Using the resulting poly A + RNA as a template, double-stranded cDNA was synthesized using the ZAP-cDNA synthesis kit (Stratagene), then, a cDNA library was prepared using the Uni-ZAP XR Cloning Kit (Stratagene) according to the method recommended by the manufacturer.

#### (2) Cloning of 5GT cDNA

The  $\lambda$  phage library obtained as described above was screened in the following manner using the p3R4 cDNA of Perilla as a probe. The filters were maintained at 42°C for 1 hour in hybridization buffer (5X SSC, 30% formamide, 50 mM sodium phosphate buffer (pH 7.0), 3% SDS 2% blocking reagent (Boehringer), 0.1% lauroylsarcosine, 80  $\mu$ g/ml of salmon sperm DNA). DIG-labeled Perilla 5GT cDNA, p3R4 cDNA, fragment was added to the hybridization solution and the filters were incubated for further 16 hours.

After washing the filters with washing solution (5X SSC 50°C, 1% SDS), the positive clones labeled with anti-DIG-alkaline phosphate were immunologically detected using 5-bromo-4-chloro-3-indolylphosphate and nitro blue tetrazolium salt according to the method described by the manufacturer (Boehringer).

As a result, seven positive clones were obtained. These cDNA were excised on plasmid pBluescript SK using the method recommended by Stratagene. When the lengths of the cDNA were investigated by agarose gel electrophoresis, insertion of a maximum length of 2.0 kb was observed.

#### (3) Determination of Nucleotide Sequence

Plasmids were extracted from the resulting clones, and the nucleotide sequences near the 3' and 5' ends of the cDNA were determined according to the dideoxy sequence method using fluorescent reagent as recommended by Perkin-Elmer with the ABI 373A sequencer (Perkin-Elmer). As a result, five of the seven clones had mutually same nucleotide sequences although the lengths of the cDNA were different. The entire nucleotide sequence of pSHGT8 was determined. Determination of nucleotide sequences was performed as described above by either using the Kilo-Sequence Deletion Kit (Takara Shuzo) to obtain a series of deleted cDNA clones, or by using an oligoprimers specific for the internal sequence of pSHGT8.

#### (4) Comparison of the Nucleotide Sequence and the Amino Acid Sequence

The cDNA inserted into pSHGT8 had the length of 2062 bp, and included an open reading frame (ORF) consisting of 1386 bp in length (including a stop codon). This sequence is shown in SEQ ID NO: 3. The amino acid sequence of this ORF had homology of 68% with the amino acid sequence of Perilla 5GT encoded by p3R4, and homology of 64% with that encoded by p3R6. In addition, it also had homology of 22 to 25% with the 3GTs of monocotyledonous and dicotyledonous plants, and homology of 21% with petunia 3RT.

#### (5) Expression in Yeast and Measurement of Enzymatic Activity

An approximately 2.0 kb DNA fragment obtained by digesting pSHGT8 with BamHI/XhoI, and an approximately 8 kb DNA fragment obtained by digesting pYE22m with BamHI/SalI were ligated, and the resulting plasmid was designated as pYHGT8. pYHGT8 was expressed in yeast cells in the same manner as Example 1, and the enzymatic activity of the protein encoded by pSHGT8 was measured. As a result, in the reaction mixture containing the crude enzyme solution of yeast transformed with pYHGT8, a product was obtained that coincided with cyanidin-3,5-

diglucoside in both retention time and absorption spectrum. Based on this observation, the pSHGT8 cDNA of Verbena was determined to code for 5GT.

Example 3 Cloning of Torenia 5GT Gene

5 (1) Preparation of cDNA Library

Petals were collected from torenia variety Summer Wave Blue (Suntory) and ground in a mortar and pestle in liquid nitrogen. RNA was extracted from the ground tissues according to a method using guanidine thiocyanate/cesium  
10 chloride, and poly A + RNA was obtained by the method recommended by the manufacturer using Oligotex (Takara Shuzo). The method using guanidine thiocyanate/cesium chloride was carried out in accordance with the method described in detail in Methods in Molecular Biology, Vol.  
15 2 (Humana Press Inc., 1984) by R. McGookin and Robert J. Slater, et al.

Using the resulting poly A + RNA as a template, double-strand cDNA was synthesized using the ZAP-cDNA synthesis kit of Strategene, then, a cDNA library was  
20 prepared using the Uni-ZAP XR Cloning Kit (Stratagene) according to the method recommended by the manufacturer.

(2) Cloning of 5GT cDNA

The  $\lambda$  phage library obtained as described above was screened in the same manner as Example 2 using the p3R4  
25 cDNA of Perilla as a probe. As a result, eight positive clones were obtained. After excision of the cDNA on plasmid pBluescript SK, the lengths of the cDNA were investigated by agarose gel electrophoresis, which revealed that a maximum length of insertion was 1.6 kb.

30 (3) Determination of Nucleotide Sequence

Plasmids were extracted from the resulting clones, and the nucleotide sequences near both 5' and 3' ends were determined in the same manner as Example 2. As a result,  
35 six of the eight clones were considered to have mutually same nucleotide sequences although the lengths of the cDNA were different. The entire nucleotide sequence of pSTGT5 cDNA was determined.



(4) Comparison of the Nucleotide Sequence and the Amino Acid Sequence

The cDNA encoded in pSTGT5 was of 1671 bp in length, and included an open reading frame (ORF) consisting of 1437 bp in length (including a stop codon). This sequence is shown in SEQ ID NO: 4. The amino acid sequence of this ORF had homology of 58% with the amino acid sequence of Perilla 5GT encoded by p3R4, and homology of 57% with that encoded by p3R6, and, homology of 57% with that encoded by Verbena pSHGT8. In addition, it also had homology of 19 to 23% with the 3GT of monocotyledonous and dicotyledonous plants, and homology of 20% with petunia 3RT.

(5) Expression of 5GT gene in Yeast

An approximately 1.6 kb DNA fragment obtained by digesting pSTGT5 with SmaI/KpnI, and an approximately 8 kb DNA fragment obtained by blunting the EcoRI-digested site of pYE22m and then digesting with KpnI were ligated, and the resulting plasmid was designated as pYTGT5. pYTGT5 was expressed in yeast cells in the same manner as Example 1, and the enzymatic activity of the protein encoded by pSTGT5 was measured. As a result, in the reaction mixture containing the crude enzyme solution of yeast transformed with pYTGT5, a product was obtained that coincided with cyanidin-3,5-diglucoside in both retention time and absorption spectrum. Based on this observation, the pSTGT5 cDNA of Torenia was determined to code for 5GT.

Example 4 Cloning of Petunia 5GT Gene

(1) Preparation of cDNA Library

A cDNA library was prepared by RNA extracted from petals of the Petunia variety Old Glory Blue in the manner described in detail by T. Holton, et al. (Plant Journal, 1993 4: 1003-1010)

(2) Cloning of 5GT cDNA

The cDNA library was screened in the same manner as Example 2 using the mixture of 5GT cDNAs of Perilla, torenia and verbena obtained in the manner described above

as probes. As a result, four positive cDNA clones were obtained and excised on plasmid pBluescript SK. The lengths of the cDNA were investigated by agarose gel electrophoresis, cDNA of a maximum length of 2.0 kb was observed.

### (3) Determination of the Nucleotide Sequence

Plasmids were extracted from the resulting clones, and the nucleotide sequence near the 5' end was determined in the same manner as Example 2. As a result, two of the four clones, pSPGT1, were appeared to code an amino acid sequence with a high degree of homology with those of 5GT from Perilla, torenia and verbena obtained thus far. Therefore, the entire nucleotide sequence of pSPGT1 was determined.

### (4) Comparison of the Nucleotide Sequence and the Amino Acid Sequence

The pSPGT1 cDNA was 2015 bp in length, and included an open reading frame (ORF) consisting of 1407 bp (including a stop codon). This sequence is shown in SEQ ID NO: 6. The amino acid sequence of this ORF had homology of 57% with that of 5GT encoded by p3R4 of Perilla, homology of 54% with that encoded by p3R6, 55% with that encoded by pSHGT8 of verbena, and 51% of that encoded by pTGT5 of torenia. In addition, it also had homology of 20 to 29% with the 3GT of monocotyledonous and dicotyledonous plants, and homology of 20% with petunia 3RT. Based on this observation, pSPGT1 cDNA obtained from petunia is considered to code for 5GT.

### Industrial Applicability

As has been described above, cDNA coding for enzymes that transfer a glycoside to the 5 position of a flavonoid originating in Perilla, verbena, torenia and petunia were cloned and their nucleotide sequences were determined. In addition, the isolated cDNAs were clearly shown to code for 5GT by the enzymatic activity of their protein expressed in yeast. Introducing of these cDNAs into a

5

CLAIMS

1. A gene coding for a protein having activity that transfers a glycoside to the 5 position of a flavonoid.

2. A gene as set forth in claim 1 that codes for a  
5 protein having an amino acid sequence described in any one  
of SEQ ID NOs: 7 through 10 or 12 and having activity that  
transfers a glycoside to the 5 position of a flavonoid, or  
a protein having an amino acid sequence modified by  
addition and/or deletion of one or more amino acids and/or  
10 substitutions by one or more other amino acids relative to  
said amino acids and maintains activity that transfers a  
glycoside to the 5 position of a flavonoid.

3. A gene as set forth in claim 1 that codes for a  
protein having an amino acid sequence that has homology of  
15 30% or more with an amino acid sequence described in any  
one of SEQ ID NOs: 7 through 10 or 12, and has activity  
that transfers a glycoside to the 5 position of a  
flavonoid.

4. A gene as set forth in claim 1 that codes for a  
20 protein having an amino acid sequence that has homology of  
50% or more with an amino acid sequence described in any  
one of SEQ ID NOs: 7 through 10 or 12, and has activity  
that transfers a glycoside to the 5 position of a  
flavonoid.

25 5. A gene as set forth in claim 1 that codes for a  
protein, wherein said gene can be hybridized under  
conditions of 5 x SCC and 50°C with all or a portion of a  
nucleotide sequence that codes for an amino acid sequence  
described in any one of SEQ ID NOs: 7 through 10 or 12,  
30 and has activity that transfers a glycoside to the 5  
position of a flavonoid.

6. A vector containing a gene as set forth in any  
one of claims 1 through 5.

7. A host transformed with a vector as set forth in  
35 claim 6.

8. A protein encoded by a gene as set forth in any  
one of claims 1 through 5.

9. A process for producing a protein comprising culturing or breeding a host as set forth in claim 7, and recovering a protein having activity that transfers a glycoside to the 5 position of a flavonoid from said host.

5 10. A plant into which is introduced a gene as set forth in any one of claims 1 through 5, or its progeny or tissue having identical properties.

11. A cut flower of the plant as set forth in claim 10 or its progeny having identical properties.

Patented September 15, 1983

ABSTRACT

The present invention provides a gene that codes for a protein having an amino acid sequence described in any of SEQ ID NOs: 7 through 10 or 12 and having activity that transfers a glycoside to the 5 position of a flavonoid, a gene that codes for a protein having a modified amino acid sequence relative to the above amino acid sequence and having activity that transfers a glycoside to the 5 position of a flavonoid, and a process for producing the above protein using said gene. This gene can be used to artificially alter the color of plants.

664280 " 5564150

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aaa atc acg ttc gtg gtg tac tcc cac ctc ttt gca tgg gcg gcc aag	388
Lys Ile Thr Phe Val Val Tyr Ser His Leu Phe Ala Trp Ala Ala Lys	
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Ala Thr Val Leu Asp Ile Phe Tyr Phe Tyr Phe Asn Gly Tyr Ser Asp	
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Glu Arg Phe Arg Ser Leu Met Lys Glu Lys Leu Glu Thr Leu Glu Gly	
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Glu Glu Lys Pro Lys Val Leu Val Asn Ser Phe Asp Ala Leu Glu Pro	
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Gly Ser Phe Val Asn Thr Thr Lys Ser Gln Met Glu Glu Ile Ala Arg	
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Glu Gly Glu Glu Val Leu Ile Ser Cys Met Glu Glu Leu Lys Arg Val	
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Gly Lys Ile Val Ser Trp Cys Ser Gln Leu Glu Val Leu Thr His Pro	
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Ser Ile Ser Phe Gly Val Pro Met Val Ala Phe Pro Gln Trp Phe Asp	
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Gln Gly Thr Asn Ala Lys Leu Met Glu Asp Val Trp Arg Thr Gly Val	
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Arg Val Arg Ala Asn Glu Glu Gly Ser Val Val Asp Gly Asp Glu Ile	
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Leu Arg Glu Ser Ala Gly Lys Trp Lys Asp Leu Ala Arg Lys Ala Met	
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Lys Arg His Ile Leu Leu Ala Thr Phe Pro Ala Gln Gly His Ile Asn  
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Pro Ser Leu Glu Phe Ala Lys Arg Leu Leu Asn Thr Gly Tyr Val Asp  
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Glu Thr Asp Pro Ser Ser Arg Ile Asp Phe Val Ala Xaa Xaa Asp Ser  
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Tyr Asp Asp Gly Leu Lys Lys Gly Asp Asp Gly Lys Asn Tyr Met Ser	
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Leu Asn Asp Ala Ala Met Gly Ser Glu Cys Tyr Asn Arg Val Ser Phe	
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Val Val Tyr Ser His Leu Phe Ser Trp Ala Ala Glu Val Ala Arg Glu	
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Val Asp Val Pro Ser Ala Leu Leu Trp Ile Glu Pro Ala Thr Val Phe	
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Asp Val Tyr Tyr Phe Tyr Phe Asn Gly Tyr Ala Asp Asp Ile Asp Ala	
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Gly Ser Asp Gln Ile Gln Leu Pro Asn Leu Pro Gln Leu Ser Lys Gln	
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Leu Met Lys Glu Lys Phe Asp Thr Leu Asp Lys Glu Pro Lys Ala Lys	
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Val Leu Ile Asn Thr Phe Asp Ala Leu Glu Thr Glu Gln Leu Lys Ala	
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Ile Asp Arg Tyr Glu Leu Ile Ser Ile Gly Pro Leu Ile Pro Ser Ser	
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Ile Phe Ser Asp Gly Asn Asp Pro Ser Ser Ser Asn Lys Ser Tyr Gly	
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Cys Phe Pro Gln Trp Phe Asp Gln Gly Thr Asn Ala Lys Met Ile Glu	
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Val Val Asp Arg Arg Glu Ile Lys Arg Cys Val Ser Glu Val Ile Lys	
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Cys	Ser	Gln	Leu	Glu	Val	Leu	Lys	His	Pro	Ser	Leu	Gly	Cys	Phe	Val	
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Met	Asp	Gly	Gly	Glu	Lys	Gly	Glu	Glu	Leu	Arg	Lys	Asn	Ala	Lys	Lys	
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 Asp Val Thr Phe Phe Thr Ser Val Tyr Ala Trp Arg Arg Met Ala Asn  
 35 40 45  
 Thr Ala Ser Ala Ala Ala Gly Asn Pro Pro Gly Leu Asp Phe Val Ala  
 50 55 60  
 Phe Ser Asp Gly Tyr Asp Asp Gly Leu Lys Pro Cys Gly Asp Gly Lys  
 65 70 75 80  
 Arg Tyr Met Ser Glu Met Lys Ala Arg Gly Ser Glu Ala Leu Arg Asn  
 85 90 95  
 Leu Leu Leu Asn Asn His Asp Val Thr Phe Val Val Tyr Ser His Leu  
 100 105 110  
 Phe Ala Trp Ala Ala Glu Val Ala Arg Glu Ser Gln Val Pro Ser Ala  
 115 120 125  
 Leu Leu Trp Val Glu Pro Ala Thr Val Leu Cys Ile Tyr Tyr Phe Tyr  
 130 135 140

Phe	Asn	Gly	Tyr	Ala	Asp	Glu	Ile	Asp	Ala	Gly	Ser	Asp	Glu	Ile	Gln
145					150					155					160
Leu	Pro	Arg	Leu	Pro	Pro	Leu	Glu	Gln	Arg	Ser	Leu	Pro	Thr	Phe	Leu
				165					170					175	
Leu	Pro	Glu	Thr	Pro	Glu	Arg	Phe	Arg	Leu	Met	Met	Lys	Glu	Lys	Leu
			180					185					190		
Glu	Thr	Leu	Asp	Gly	Glu	Glu	Lys	Ala	Lys	Val	Leu	Val	Asn	Thr	Phe
		195					200					205			
Asp	Ala	Leu	Glu	Pro	Asp	Ala	Leu	Thr	Ala	Ile	Asp	Arg	Tyr	Glu	Leu
	210					215					220				
Ile	Gly	Ile	Gly	Pro	Leu	Ile	Pro	Ser	Ala	Phe	Leu	Asp	Gly	Gly	Asp
225					230					235					240
Pro	Ser	Glu	Thr	Ser	Tyr	Gly	Gly	Asp	Leu	Phe	Glu	Lys	Ser	Glu	Glu
			245					250						255	
Asn	Asn	Cys	Val	Glu	Trp	Leu	Asp	Thr	Lys	Pro	Lys	Ser	Ser	Val	Val
		260					265						270		
Tyr	Val	Ser	Phe	Gly	Ser	Val	Leu	Arg	Phe	Pro	Lys	Ala	Gln	Met	Glu
	275						280					285			
Glu	Ile	Gly	Lys	Gly	Leu	Leu	Ala	Cys	Gly	Arg	Pro	Phe	Leu	Trp	Met
	290					295					300				
Ile	Arg	Glu	Gln	Lys	Asn	Asp	Asp	Gly	Glu	Glu	Glu	Glu	Glu	Glu	Leu
305					310					315					320
Ser	Cys	Ile	Gly	Glu	Leu	Lys	Lys	Met	Gly	Lys	Ile	Val	Ser	Trp	Cys
			325						330					335	
Ser	Gln	Leu	Glu	Val	Leu	Ala	His	Pro	Ala	Leu	Gly	Cys	Phe	Val	Thr
		340						345					350		
His	Cys	Gly	Trp	Asn	Ser	Ala	Val	Glu	Ser	Leu	Ser	Cys	Gly	Val	Pro
	355						360					365			
Val	Val	Ala	Val	Pro	Gln	Trp	Phe	Asp	Gln	Thr	Thr	Asn	Ala	Lys	Leu
	370					375					380				
Ile	Glu	Asp	Ala	Trp	Gly	Thr	Gly	Val	Arg	Val	Arg	Met	Asn	Glu	Gly
385					390					395					400
Gly	Gly	Val	Asp	Gly	Ser	Glu	Ile	Glu	Arg	Cys	Val	Glu	Met	Val	Met
			405						410					415	



Asp Gly Gly Glu Lys Ser Lys Leu Val Arg Glu Asn Ala Ile Lys Trp  
                     420                                    425                                    430  
 Lys Thr Leu Ala Arg Glu Ala Met Gly Glu Asp Gly Ser Ser Leu Lys  
                     435                                    440                                    445  
 Asn Leu Asn Ala Phe Leu His Gln Val Ala Arg Ala  
                     450                                    455                                    460  
  
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 Ile Asn Pro Ala Leu Gln Phe Ala Lys Arg Leu Leu Lys Ala Gly Thr  
                                     20                                    25                                    30  
 Asp Val Thr Phe Phe Thr Ser Val Tyr Ala Trp Arg Arg Met Ala Asn  
                     35                                    40                                    45  
 Thr Ala Ser Ala Ala Ala Gly Asn Pro Pro Gly Leu Asp Phe Val Ala  
                     50                                    55                                    60  
 Phe Ser Asp Gly Tyr Asp Asp Gly Leu Lys Pro Gly Gly Asp Gly Lys  
   65                                    70                                    75                                    80  
 Arg Tyr Met Ser Glu Met Lys Ala Arg Gly Ser Glu Ala Leu Arg Asn  
                                     85                                    90                                    95  
 Leu Leu Leu Asn Asn Asp Asp Val Thr Phe Val Val Tyr Ser His Leu  
                     100                                    105                                    110  
 Phe Ala Trp Ala Ala Glu Val Ala Arg Leu Ser His Val Pro Thr Ala  
                     115                                    120                                    125  
 Leu Leu Trp Val Glu Pro Ala Thr Val Leu Cys Ile Tyr His Phe Tyr  
                     130                                    135                                    140  
 Phe Asn Gly Tyr Ala Asp Glu Ile Asp Ala Gly Ser Asn Glu Ile Gln  
   145                                    150                                    155                                    160  
 Leu Pro Arg Leu Pro Ser Leu Glu Gln Arg Ser Leu Pro Thr Phe Leu  
                                     165                                    170                                    175  
 Leu Pro Ala Thr Pro Glu Arg Phe Arg Leu Met Met Lys Glu Lys Leu  
                     180                                    185                                    190

Glu Thr Leu Asp Gly Glu Glu Lys Ala Lys Val Leu Val Asn Thr Phe		
195	200	205
Asp Ala Leu Glu Pro Asp Ala Leu Thr Ala Ile Asp Arg Tyr Glu Leu		
210	215	220
Ile Gly Ile Gly Pro Leu Ile Pro Ser Ala Phe Leu Asp Gly Glu Asp		
225	230	235
Pro Ser Glu Thr Ser Tyr Gly Gly Asp Leu Phe Glu Lys Ser Glu Glu		
245	250	255
Asn Asn Cys Val Glu Trp Leu Asn Ser Lys Pro Lys Ser Ser Val Val		
260	265	270
Tyr Val Ser Phe Gly Ser Val Leu Arg Phe Pro Lys Ala Gln Met Glu		
275	280	285
Glu Ile Gly Lys Gly Leu Leu Ala Cys Gly Arg Pro Phe Leu Trp Met		
290	295	300
Ile Arg Glu Gln Lys Asn Asp Asp Gly Glu Glu Glu Glu Glu Glu		
305	310	315
Glu Leu Ser Cys Ile Gly Glu Leu Lys Lys Met Gly Lys Ile Val Ser		
325	330	335
Trp Cys Ser Gln Leu Glu Val Leu Ala His Pro Ala Leu Gly Cys Phe		
340	345	350
Val Thr His Cys Gly Trp Asn Ser Ala Val Glu Ser Leu Ser Cys Gly		
355	360	365
Ile Pro Val Val Ala Val Pro Gln Trp Phe Asp Gln Thr Thr Asn Ala		
370	375	380
Lys Leu Ile Glu Asp Ala Trp Gly Thr Gly Val Arg Val Arg Met Asn		
385	390	395
Glu Gly Gly Gly Val Asp Gly Cys Glu Ile Glu Arg Cys Val Glu Met		
405	410	415
Val Met Asp Gly Gly Asp Lys Thr Lys Leu Val Arg Glu Asn Ala Ile		
420	425	430
Lys Trp Lys Thr Leu Ala Arg Gln Ala Met Gly		
435	440	443

<210> 9

<211> 461

<212> PRT

<213> Verbena hybrida

<400> 9

Met	Ser	Arg	Ala	His	Val	Leu	Leu	Ala	Thr	Phe	Pro	Ala	Gln	Gly	His
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Ile	Asn	Pro	Ala	Leu	Gln	Phe	Ala	Lys	Arg	Leu	Ala	Asn	Ala	Asp	Ile
			20					25					30		
Gln	Val	Thr	Phe	Phe	Thr	Ser	Val	Tyr	Ala	Trp	Arg	Arg	Met	Ser	Arg
		35					40					45			
Thr	Ala	Ala	Gly	Ser	Asn	Gly	Leu	Ile	Asn	Phe	Val	Ser	Phe	Ser	Asp
	50					55					60				
Gly	Tyr	Asp	Asp	Gly	Leu	Gln	Pro	Gly	Asp	Asp	Gly	Lys	Asn	Tyr	Met
65					70				75					80	
Ser	Glu	Met	Lys	Ser	Arg	Gly	Ile	Lys	Ala	Leu	Ser	Asp	Thr	Leu	Ala
				85				90						95	
Ala	Asn	Asn	Val	Asp	Gln	Lys	Ser	Ser	Lys	Ile	Thr	Phe	Val	Val	Tyr
			100					105					110		
Ser	His	Leu	Phe	Ala	Trp	Ala	Ala	Lys	Val	Ala	Arg	Glu	Phe	His	Leu
		115					120						125		
Arg	Ser	Ala	Leu	Leu	Trp	Ile	Glu	Pro	Ala	Thr	Val	Leu	Asp	Ile	Phe
	130						135					140			
Tyr	Phe	Tyr	Phe	Asn	Gly	Tyr	Ser	Asp	Glu	Ile	Asp	Ala	Gly	Ser	Asp
145					150					155				160	
Ala	Ile	His	Leu	Pro	Gly	Gly	Leu	Pro	Val	Leu	Ala	Gln	Arg	Asp	Leu
			165					170					175		
Pro	Ser	Phe	Leu	Leu	Pro	Ser	Thr	His	Glu	Arg	Phe	Arg	Ser	Leu	Met
		180						185					190		
Lys	Glu	Lys	Leu	Glu	Thr	Leu	Glu	Gly	Glu	Glu	Lys	Pro	Lys	Val	Leu
	195						200					205			
Val	Asn	Ser	Phe	Asp	Ala	Leu	Glu	Pro	Asp	Ala	Leu	Lys	Ala	Ile	Asp
	210						215					220			
Lys	Tyr	Glu	Met	Ile	Ala	Ile	Gly	Pro	Leu	Ile	Pro	Ser	Ala	Phe	Leu
225					230					235				240	
Asp	Gly	Lys	Asp	Pro	Ser	Asp	Arg	Ser	Phe	Gly	Gly	Asp	Leu	Phe	Glu
			245					250					255		
Lys	Gly	Ser	Asn	Asp	Asp	Asp	Cys	Leu	Glu	Trp	Leu	Ser	Thr	Asn	Pro
		260						265					270		

Arg	Ser	Ser	Val	Val	Tyr	Val	Ser	Phe	Gly	Ser	Phe	Val	Asn	Thr	Thr	
275					280					285						
Lys	Ser	Gln	Met	Glu	Glu	Ile	Ala	Arg	Gly	Leu	Leu	Asp	Cys	Gly	Arg	
290					295					300						
Pro	Phe	Leu	Trp	Val	Val	Arg	Val	Asn	Glu	Gly	Glu	Glu	Val	Leu	Ile	
305					310					315					320	
Ser	Cys	Met	Glu	Glu	Leu	Lys	Arg	Val	Gly	Lys	Ile	Val	Ser	Trp	Cys	
325					330					335						
Ser	Gln	Leu	Glu	Val	Leu	Thr	His	Pro	Ser	Leu	Gly	Cys	Phe	Val	Thr	
340					345					350						
His	Cys	Gly	Trp	Asn	Ser	Thr	Leu	Glu	Ser	Ile	Ser	Phe	Gly	Val	Pro	
355					360					365						
Met	Val	Ala	Phe	Pro	Gln	Trp	Phe	Asp	Gln	Gly	Thr	Asn	Ala	Lys	Leu	
370					375					380						
Met	Glu	Asp	Val	Trp	Arg	Thr	Gly	Val	Arg	Val	Arg	Ala	Asn	Glu	Glu	
385					390					395					400	
Gly	Ser	Val	Val	Asp	Gly	Asp	Glu	Ile	Arg	Arg	Cys	Ile	Glu	Glu	Val	
405					410					415						
Met	Asp	Gly	Gly	Glu	Lys	Ser	Arg	Lys	Leu	Arg	Glu	Ser	Ala	Gly	Lys	
420					425					430						
Trp	Lys	Asp	Leu	Ala	Arg	Lys	Ala	Met	Glu	Glu	Asp	Gly	Ser	Ser	Val	
435					440					445						
Asn	Asn	Leu	Lys	Val	Phe	Leu	Asp	Glu	Val	Val	Gly	Ile				
450					455					460 461						

<210> 10

<211> 478

<212> PRT

<213> *Torenia hybrida*

<220>

<221>

<222>

<223> Xaa (64) is Cys or Phe, Xaa (65) is Ser or Pro.

<400> 10

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                   20                                  25                                  30  
 Tyr Val Asp Gln Val Thr Phe Phe Thr Ser Val Tyr Ala Leu Arg Arg  
                   35                                  40                                  45  
 Met Arg Phe Glu Thr Asp Pro Ser Ser Arg Ile Asp Phe Val Ala Xaa  
                   50                                  55                                  60  
 Xaa Asp Ser Tyr Asp Asp Gly Leu Lys Lys Gly Asp Asp Gly Lys Asn  
                   65                                  70                                  75                                  80  
 Tyr Met Ser Glu Met Arg Lys Arg Gly Thr Lys Ala Leu Lys Asp Thr  
                                   85                                  90                                  95  
 Leu Ile Lys Leu Asn Asp Ala Ala Met Gly Ser Glu Cys Tyr Asn Arg  
                   100                                  105                                  110  
 Val Ser Phe Val Val Tyr Ser His Leu Phe Ser Trp Ala Ala Glu Val  
                   115                                  120                                  125  
 Ala Arg Glu Val Asp Val Pro Ser Ala Leu Leu Trp Ile Glu Pro Ala  
                   130                                  135                                  140  
 Thr Val Phe Asp Val Tyr Tyr Phe Tyr Phe Asn Gly Tyr Ala Asp Asp  
                   145                                  150                                  155                                  160  
 Ile Asp Ala Gly Ser Asp Gln Ile Gln Leu Pro Asn Leu Pro Gln Leu  
                                   165                                  170                                  175  
 Ser Lys Gln Asp Leu Pro Ser Phe Leu Leu Pro Ser Ser Pro Ala Arg  
                   180                                  185                                  190  
 Phe Arg Thr Leu Met Lys Glu Lys Phe Asp Thr Leu Asp Lys Glu Pro  
                   195                                  200                                  205  
 Lys Ala Lys Val Leu Ile Asn Thr Phe Asp Ala Leu Glu Thr Glu Gln  
                   210                                  215                                  220  
 Leu Lys Ala Ile Asp Arg Tyr Glu Leu Ile Ser Ile Gly Pro Leu Ile  
                   225                                  230                                  235                                  240  
  
 Pro Ser Ser Ile Phe Ser Asp Gly Asn Asp Pro Ser Ser Ser Asn Lys  
                   245                                  250                                  255  
 Ser Tyr Gly Gly Asp Leu Phe Arg Lys Ala Asp Glu Thr Tyr Met Asp  
                   260                                  265                                  270  
 Trp Leu Asn Ser Lys Pro Glu Ser Ser Val Val Tyr Val Ser Phe Gly  
                   275                                  280                                  285

Ser Leu Leu Arg Leu Pro Lys Pro Gln Met Glu Glu Ile Ala Ile Gly  
 290 295 300  
 Leu Ser Asp Thr Lys Ser Pro Val Leu Trp Val Ile Arg Arg Asn Glu  
 305 310 315 320  
 Glu Gly Asp Glu Gln Glu Gln Ala Glu Glu Glu Glu Lys Leu Leu Ser  
 325 330 335  
 Phe Phe Asp Arg His Gly Thr Glu Arg Leu Gly Lys Ile Val Thr Trp  
 340 345 350  
 Cys Ser Gln Leu Asp Val Leu Thr His Lys Ser Val Gly Cys Phe Val  
 355 360 365  
 Thr His Cys Gly Trp Asn Ser Ala Ile Glu Ser Leu Ala Cys Gly Val  
 370 375 380  
 Pro Val Val Cys Phe Pro Gln Trp Phe Asp Gln Gly Thr Asn Ala Lys  
 385 390 395 400  
 Met Ile Glu Asp Val Trp Arg Ser Gly Val Arg Val Arg Val Asn Glu  
 405 410 415  
 Glu Gly Gly Val Val Asp Arg Arg Glu Ile Lys Arg Cys Val Ser Glu  
 420 425 430  
 Val Ile Lys Ser Arg Glu Leu Arg Glu Ser Ala Met Met Trp Lys Gly  
 435 440 445  
 Leu Ala Lys Glu Ala Met Asp Glu Glu Arg Gly Ser Ser Met Asn Asn  
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 <213> *Perilla frutescens*  
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 Ala Ile Met Thr Phe Ser Ala Ser Leu Val Ser Thr Thr Val Asp Ala  
 35 40 45

Pro Leu Thr Met Ser Phe Thr Thr Tyr Thr Val Val Ala Leu Leu Tyr  
 50 55 60  
 Gly Thr Ile Leu Leu Tyr Arg Arg His Lys Phe Leu Val Pro Trp Tyr  
 65 70 75 80  
 Trp Tyr Ala Leu Leu Gly Phe Val Asp Val His Gly Asn Tyr Leu Val  
 85 90 95  
 Asn Lys Ala Phe Glu Leu Thr Ser Ile Thr Ser Val Ser Ile Leu Asp  
 100 105 110  
 Cys Trp Thr Ile Val Trp Ser Ile Ile Phe Thr Trp Met Phe Leu Gly  
 115 120 125  
 Thr Lys Tyr Ser Val Tyr Gln Phe Val Gly Ala Ala Ile Cys Val Gly  
 130 135 140  
 Gly Leu Leu Leu Val Leu Leu Ser Asp Ser Gly Val Thr Ala Ala Gly  
 145 150 155 160  
 Ser Asn Pro Leu Leu Gly Asp Phe Leu Val Ile Thr Gly Ser Ile Leu  
 165 170 175  
 Phe Thr Leu Ser Thr Val Gly Gln Glu Tyr Cys Val Lys Arg Lys Asp  
 180 185 190  
 Arg Ile Glu Val Val Ala Met Ile Gly Val Phe Gly Met Leu Ile Ser  
 195 200 205  
 Ala Thr Glu Ile Thr Val Leu Glu Arg Asn Ala Leu Ser Ser Met Gln  
 210 215 220  
 Trp Ser Thr Gly Leu Leu Ala Ala Tyr Val Val Tyr Ala Leu Ser Ser  
 225 230 235 240  
 Phe Leu Phe Cys Thr Leu Thr Pro Phe Leu Leu Lys Met Ser Gly Ala  
 245 250 255  
 Ala Phe Phe Asn Leu Ser Met Leu Thr Ser Asp Met Trp Ala Val Ala  
 260 265 270  
 Ile Arg Thr Phe Ile Tyr Asn Gln Glu Val Asp Trp Leu Tyr Tyr Leu  
 275 280 285  
 Ala Phe Cys Leu Val Val Val Gly Ile Phe Ile Tyr Thr Lys Thr Glu  
 290 295 300  
 Lys Asp Pro Asn Asn Thr Arg Ala Leu Glu Asn Gly Asn Leu Asp His  
 305 310 315 320  
 Glu Tyr Ser Leu Leu Glu Asp Gln Asp Asp Thr Pro Arg Lys Pro  
 325 330 335  
 <210> 12  
 <211> 468

<212> PRT

<213> Petunia hybrida

<400> 12

Met Val Gln Pro His Val Ile Leu Thr Thr Phe Pro Ala Gln Gly His  
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Ile Asn Pro Ala Leu Gln Phe Ala Lys Asn Leu Val Lys Met Gly Ile  
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Glu Val Thr Phe Ser Thr Ser Ile Tyr Ala Gln Ser Arg Met Asp Glu  
35 40 45  
Lys Ser Ile Leu Asn Ala Pro Lys Gly Leu Asn Phe Ile Pro Phe Ser  
50 55 60  
Asp Gly Phe Asp Glu Gly Phe Asp His Ser Lys Asp Pro Val Phe Tyr  
65 70 75 80  
Met Ser Gln Leu Arg Lys Cys Gly Ser Glu Thr Val Lys Lys Ile Ile  
85 90 95  
Leu Thr Cys Ser Glu Asn Gly Gln Pro Ile Thr Cys Leu Leu Tyr Ser  
100 105 110  
Ile Phe Leu Pro Trp Ala Ala Glu Val Ala Arg Glu Val His Ile Pro  
115 120 125  
Ser Ala Leu Leu Trp Ser Gln Pro Ala Thr Ile Leu Asp Ile Tyr Tyr  
130 135 140  
Phe Asn Phe His Gly Tyr Glu Lys Ala Met Ala Asn Glu Ser Asn Asp  
145 150 155 160  
Pro Asn Trp Ser Ile Gln Leu Pro Gly Leu Pro Leu Leu Glu Thr Arg  
165 170 175  
Asp Leu Pro Ser Phe Leu Leu Pro Tyr Gly Ala Lys Gly Ser Leu Arg  
180 185 190  
Val Ala Leu Pro Pro Phe Lys Glu Leu Ile Asp Thr Leu Asp Ala Glu  
195 200 205  
Thr Thr Pro Lys Ile Leu Val Asn Thr Phe Asp Glu Leu Glu Pro Glu  
210 215 220  
Ala Leu Asn Ala Ile Glu Gly Tyr Lys Phe Tyr Gly Ile Gly Pro Leu  
225 230 235 240  
Ile Pro Ser Ala Phe Leu Gly Gly Asn Asp Pro Leu Asp Ala Ser Phe  
245 250 255  
Gly Gly Asp Leu Phe Gln Asn Ser Asn Asp Tyr Met Glu Trp Leu Asn  
260 265 270



Ser Lys Pro Asn Ser Ser Val Val Tyr Ile Ser Phe Gly Ser Leu Met  
 275 280 285  
 Asn Pro Ser Ile Ser Gln Met Glu Glu Ile Ser Lys Gly Leu Ile Asp  
 290 295 300  
 Ile Gly Arg Pro Phe Leu Trp Val Ile Lys Glu Asn Glu Lys Gly Lys  
 305 310 315 320  
 Glu Glu Glu Asn Lys Lys Leu Gly Cys Ile Glu Glu Leu Glu Lys Ile  
 325 330 335  
 Gly Lys Ile Val Pro Trp Cys Ser Gln Leu Glu Val Leu Lys His Pro  
 340 345 350  
 Ser Leu Gly Cys Phe Val Ser His Cys Gly Trp Asn Ser Ala Leu Glu  
 355 360 365  
 Ser Leu Ala Cys Gly Val Pro Val Val Ala Phe Pro Gln Trp Thr Asp  
 370 375 380  
 Gln Met Thr Asn Ala Lys Gln Val Glu Asp Val Trp Lys Ser Gly Val  
 385 390 395 400  
 Arg Val Arg Ile Asn Glu Asp Gly Val Val Glu Ser Glu Glu Ile Lys  
 405 410 415  
 Arg Cys Ile Glu Leu Val Met Asp Gly Gly Glu Lys Gly Glu Glu Leu  
 420 425 430  
 Arg Lys Asn Ala Lys Lys Trp Lys Glu Leu Ala Arg Glu Ala Val Lys  
 435 440 445  
 Glu Gly Gly Ser Ser His Lys Asn Leu Lys Ala Phe Ile Asp Asp Val  
 450 455 460  
 Ala Lys Gly Phe  
 465 468

# Sequence

Sequence ID No.: 1

Sequence length: 1507

Sequence type: Nucleic acid

Number of strands: Double-strand

Topology: Straight chain

Source:

Biological name: Perilla (Perilla frutescens)

Tissue type: Leaf

Direct source:

Library name: cDNA library

Clone name: p3R4

Sequence:

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      Met Val Arg Arg Arg Val Leu Leu Ala Thr Phe
              1              5              10
CCT GCG CAA GGC CAC ATA AAT CCC GCC CTC CAA TTC GCC AAG AGA CTC      97
Pro Ala Gln Gly His Ile Asn Pro Ala Leu Gln Phe Ala Lys Arg Leu
              15              20              25
CTA AAA GCC GGC ACT GAC GTC ACA TTT TTC ACG AGC GTT TAT GCA TGG      145
Leu Lys Ala Gly Thr Asp Val Thr Phe Phe Thr Ser Val Tyr Ala Trp
              30              35              40
CGC CGC ATG GCC AAC ACA GCC TCC GCC GCT GCC GGA AAC CCA CCG GGC      193
Arg Arg Met Ala Asn Thr Ala Ser Ala Ala Ala Gly Asn Pro Pro Gly
              45              50              55
CTC GAC TTC GTG GCG TTC TCC GAC GGC TAC GAC GAC GGG CTG AAG CCC      241
Leu Asp Phe Val Ala Phe Ser Asp Gly Tyr Asp Asp Gly Leu Lys Pro
              60              65              70              75
TGC GGC GAC GGG AAG CGC TAC ATG TCC GAG ATG AAA GCC CGC GGC TCC      289
Cys Gly Asp Gly Lys Arg Tyr Met Ser Glu Met Lys Ala Arg Gly Ser
              80              85              90
GAG GCC TTA AGA AAC CTC CTT CTC AAC AAC CAC GAC GTC ACG TTC GTC      337
Glu Ala Leu Arg Asn Leu Leu Leu Asn Asn His Asp Val Thr Phe Val
              95              100              105
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GenBank Entry

GTC TAC TCC CAC CTC TTT GCA TGG GCG GCG GAG GTG GCG CGT GAG TCC	385
Val Tyr Ser His Leu Phe Ala Trp Ala Ala Glu Val Ala Arg Glu Ser	
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CAG GTC CCG AGC GCC CTT CTC TGG GTC GAG CCC GCC ACC GTG CTG TGC	433
Gln Val Pro Ser Ala Leu Leu Trp Val Glu Pro Ala Thr Val Leu Cys	
125 130 135	
ATA TAT TAC TTC TAC TTC AAC GGC TAC GCA GAC GAG ATC GAC GCC GGT	481
Ile Tyr Tyr Phe Tyr Phe Asn Gly Tyr Ala Asp Glu Ile Asp Ala Gly	
140 145 150 155	
TCC GAC GAA ATT CAG CTC CCT CGG CTT CCA CCC CTG GAG CAG CGC AGT	529
Ser Asp Glu Ile Gln Leu Pro Arg Leu Pro Pro Leu Glu Gln Arg Ser	
160 165 170	
CTT CCG ACC TTT CTG CTG CCG GAG ACA CCG GAG AGA TTC CGG TTG ATG	577
Leu Pro Thr Phe Leu Leu Pro Glu Thr Pro Glu Arg Phe Arg Leu Met	
175 180 185	
ATG AAG GAG AAG CTG GAA ACT TTA GAC GGT GAA GAG AAG GCG AAA GTG	625
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190 195 200	
TTG GTG AAC ACG TTT GAT GCG TTG GAG CCC GAT GCA CTC ACG GCT ATT	673
Leu Val Asn Thr Phe Asp Ala Leu Glu Pro Asp Ala Leu Thr Ala Ile	
205 210 215	
GAT AGG TAT GAG TTG ATC GGG ATC GGG CCG TTG ATT CCC TCC GCC TTC	721
Asp Arg Tyr Glu Leu Ile Gly Ile Gly Pro Leu Ile Pro Ser Ala Phe	
220 225 230 235	
TTG GAC GGC GGA GAT CCC TCC GAA ACG TCT TAC GGC GGC GAT CTT TTC	769
Leu Asp Gly Gly Asp Pro Ser Glu Thr Ser Tyr Gly Gly Asp Leu Phe	
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GAA AAA TCG GAG GAG AAT AAC TGC GTG GAG TGG TTG GAC ACG AAG CCG	817
Glu Lys Ser Glu Glu Asn Asn Cys Val Glu Trp Leu Asp Thr Lys Pro	
255 260 265	
AAA TCT TCG GTG GTG TAT GTG TCG TTT GGG AGC GTT TTG AGG TTT CCA	865
Lys Ser Ser Val Val Tyr Val Ser Phe Gly Ser Val Leu Arg Phe Pro	
270 275 280	
AAG GCA CAA ATG GAA GAG ATT GGG AAA GGG CTA TTA GCC TGC GGA AGG	913
Lys Ala Gln Met Glu Glu Ile Gly Lys Gly Leu Leu Ala Cys Gly Arg	
285 290 295	

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CCG TTT TTA TGG ATG ATA CGA GAA CAG AAG AAT GAC GAC GGC GAA GAA      961
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GAA GAA GAA GAG TTG AGT TGC ATT GGG GAA TTG AAA AAA ATG GGG AAA      1009
Glu Glu Glu Glu Leu Ser Cys Ile Gly Glu Leu Lys Lys Met Gly Lys
                      320                      325                      330
ATA GTT TCG TGG TGC TCG CAG TTG GAG GTT CTG GCG CAC CCT GCG TTG      1057
Ile Val Ser Trp Cys Ser Gln Leu Glu Val Leu Ala His Pro Ala Leu
                      335                      340                      345
GGA TGT TTC GTG ACG CAT TGT GGG TGG AAC TCG GCT GTG GAG AGC TTG      1105
Gly Cys Phe Val Thr His Cys Gly Trp Asn Ser Ala Val Glu Ser Leu
                      350                      355                      360
AGT TGC GGG GTT CCG GTG GTG GCG GTG CCG CAG TGG TTT GAT CAG ACG      1153
Ser Cys Gly Val Pro Val Val Ala Val Pro Gln Trp Phe Asp Gln Thr
                      365                      370                      375
ACG AAT GCG AAG CTG ATT GAG GAT GCG TGG GGG ACA GGG GTG AGA GTG      1201
Thr Asn Ala Lys Leu Ile Glu Asp Ala Trp Gly Thr Gly Val Arg Val
380                      385                      390                      395
AGA ATG AAT GAA GGG GGT GGG GTT GAT GGA TCT GAG ATA GAG AGG TGT      1249
Arg Met Asn Glu Gly Gly Gly Val Asp Gly Ser Glu Ile Glu Arg Cys
                      400                      405                      410
GTG GAG ATG GTG ATG GAT GGG GGT GAG AAG AGC AAA CTA GTG AGA GAA      1297
Val Glu Met Val Met Asp Gly Gly Glu Lys Ser Lys Leu Val Arg Glu
                      415                      420                      425
AAT GCC ATA AAA TGG AAG ACT TTG GCC AGA GAA GCC ATG GGA GAG GAT      1345
Asn Ala Ile Lys Trp Lys Thr Leu Ala Arg Glu Ala Met Gly Glu Asp
                      430                      435                      440
GGA TCT TCA CTC AAG AAT CTC AAC GCC TTT CTT CAT CAA GTT GCA CGT      1393
Gly Ser Ser Leu Lys Asn Leu Asn Ala Phe Leu His Gln Val Ala Arg
                      445                      450                      455
GCT TAATACACAA AATGGCTTTC CACTTTTAAT CTA CTCAAAC ACCGGTTCAA      1446
Ala
460                                                                1507
ATAAATATCC CCTTCCACTT CTTTCTATTT CACTATCACA TTTATAATTT TAGTAACAAA 1506
A

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Sequence ID No.: 2

Sequence length: 1470

Sequence type: Nucleic acid

Number of strands: Double-strand

Topology: Straight chain

Source:

Biological name: Perilla (Perilla frutescens)

Tissue type: Leaf

Direct source:

Library name: cDNA library

Clone name: p3R6

Sequence:

```
ACCAAACCAA AACAAAATTT CCACAAAA ATG GTC CGC CGC CGC GTG CTG CTA      48
                                Met Val Arg Arg Arg Val Leu Leu
                                1           5

GCA ACG TTT CCG GCG CAA GGC CAC ATA AAT CCC GCC CTC CAA TTC GCC      96
Ala Thr Phe Pro Ala Gln Gly His Ile Asn Pro Ala Leu Gln Phe Ala
    10           15           20

AAG AGA CTC CTA AAA GCC GGC ACT GAC GTC ACG TTT TTC ACG AGC GTT      144
Lys Arg Leu Leu Lys Ala Gly Thr Asp Val Thr Phe Phe Thr Ser Val
    25           30           35           40

TAT GCA TGG CGC CGC ATG GCC AAC ACA GCC TCC GCC GCT GCC GGA AAC      192
Tyr Ala Trp Arg Arg Met Ala Asn Thr Ala Ser Ala Ala Ala Gly Asn
           45           50           55

CCA CCG GGC CTC GAC TTC GTG GCG TTC TCC GAC GGC TAC GAC GAC GGG      240
Pro Pro Gly Leu Asp Phe Val Ala Phe Ser Asp Gly Tyr Asp Asp Gly
           60           65           70

CTG AAG CCC GGC GGC GAC GGG AAG CGC TAC ATG TCC GAG ATG AAA GCC      288
Leu Lys Pro Gly Gly Asp Gly Lys Arg Tyr Met Ser Glu Met Lys Ala
           75           80           85

CGC GGC TCC GAG GCC TTA AGA AAC CTC CTT CTC AAC AAC GAC GAC GTC      336
Arg Gly Ser Glu Ala Leu Arg Asn Leu Leu Leu Asn Asn Asp Asp Val
           90           95           100

ACT TTC GTC GTC TAC TCC CAC CTC TTT GCA TGG GCG GCG GAG GTG GCG      384
Thr Phe Val Val Tyr Ser His Leu Phe Ala Trp Ala Ala Glu Val Ala
105           110           115           120
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CGT TTG TCC CAC GTC CCG ACC GCC CTT CTC TGG GTC GAG CCC GCC ACC	432
Arg Leu Ser His Val Pro Thr Ala Leu Leu Trp Val Glu Pro Ala Thr	
125 130 135	
GTG CTG TGC ATA TAC CAC TTC TAC TTC AAC GGC TAC GCA GAC GAG ATC	480
Val Leu Cys Ile Tyr His Phe Tyr Phe Asn Gly Tyr Ala Asp Glu Ile	
140 145 150	
GAC GCC GGT TCC AAT GAA ATT CAG CTC CCT CGG CTT CCA TCC CTG GAG	528
Asp Ala Gly Ser Asn Glu Ile Gln Leu Pro Arg Leu Pro Ser Leu Glu	
155 160 165	
CAG CGC AGT CTT CCG ACG TTT CTG CTG CCT GCG ACG CCG GAG AGA TTC	576
Gln Arg Ser Leu Pro Thr Phe Leu Leu Pro Ala Thr Pro Glu Arg Phe	
170 175 180	
CGG TTG ATG ATG AAG GAG AAG CTG GAA ACT TTA GAC GGT GAA GAG AAG	624
Arg Leu Met Met Lys Glu Lys Leu Glu Thr Leu Asp Gly Glu Glu Lys	
185 190 195 200	
GCG AAA GTA TTG GTG AAC ACG TTT GAT GCG TTG GAG CCC GAT GCA CTC	672
Ala Lys Val Leu Val Asn Thr Phe Asp Ala Leu Glu Pro Asp Ala Leu	
205 210 215	
ACG GCT ATT GAT AGG TAT GAG TTG ATC GGG ATC GGG CCG TTG ATT CCC	720
Thr Ala Ile Asp Arg Tyr Glu Leu Ile Gly Ile Gly Pro Leu Ile Pro	
220 225 230	
TCC GCC TTC TTG GAC GGC GAA GAT CCC TCC GAA ACG TCT TAC GGC GGC	768
Ser Ala Phe Leu Asp Gly Glu Asp Pro Ser Glu Thr Ser Tyr Gly Gly	
235 240 245	
GAT CTT TTC GAA AAA TCG GAG GAG AAT AAC TGC GTG GAG TGG TTG AAC	816
Asp Leu Phe Glu Lys Ser Glu Glu Asn Asn Cys Val Glu Trp Leu Asn	
250 255 260	
TCG AAG CCG AAA TCT TCG GTG GTG TAT GTG TCG TTT GGG AGC GTT TTG	864
Ser Lys Pro Lys Ser Ser Val Val Tyr Val Ser Phe Gly Ser Val Leu	
265 270 275 280	
AGG TTT CCA AAG GCA CAA ATG GAA GAG ATT GGG AAA GGG CTA TTA GCC	912
Arg Phe Pro Lys Ala Gln Met Glu Glu Ile Gly Lys Gly Leu Leu Ala	
285 290 295	
TGC GGA AGG CCC TTT TTA TGG ATG ATA CGA GAA CAG AAG AAT GAC GAC	960
Cys Gly Arg Pro Phe Leu Trp Met Ile Arg Glu Gln Lys Asn Asp Asp	
300 305 310	

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GGC GAA GAA GAA GAA GAA GAA GAA GAG TTG AGT TGC ATT GGG GAA TTG 1008
Gly Glu Glu Glu Glu Glu Glu Glu Glu Glu Leu Ser Cys Ile Gly Glu Leu
      315              320              325
AAA AAA ATG GGG AAA ATA GTG TCG TGG TGC TCG CAG TTG GAG GTT CTG 1056
Lys Lys Met Gly Lys Ile Val Ser Trp Cys Ser Gln Leu Glu Val Leu
      330              335              340
GCG CAC CCT GCG TTG GGA TGT TTC GTG ACG CAT TGT GGG TGG AAC TCG 1104
Ala His Pro Ala Leu Gly Cys Phe Val Thr His Cys Gly Trp Asn Ser
345              350              355              360
GCT GTG GAG AGC TTG AGT TGC GGG ATT CCG GTG GTG GCG GTG CCG CAG 1152
Ala Val Glu Ser Leu Ser Cys Gly Ile Pro Val Val Ala Val Pro Gln
      365              370              375
TGG TTT GAT CAG ACG ACG AAT GCG AAG CTG ATT GAG GAT GCG TGG GGG 1200
Trp Phe Asp Gln Thr Thr Asn Ala Lys Leu Ile Glu Asp Ala Trp Gly
      380              385              390
ACA GGG GTG AGA GTG AGA ATG AAT GAA GGG GGT GGG GTT GAT GGA TGT 1248
Thr Gly Val Arg Val Arg Met Asn Glu Gly Gly Gly Val Asp Gly Cys
      395              400              405
GAG ATA GAA AGG TGT GTG GAG ATG GTG ATG GAT GGG GGT GAC AAG ACC 1296
Glu Ile Glu Arg Cys Val Glu Met Val Met Asp Gly Gly Asp Lys Thr
      410              415              420
AAA CTA GTG AGA GAA AAT GCC ATC AAA TGG AAG ACT TTG GCC AGA CAA 1344
Lys Leu Val Arg Glu Asn Ala Ile Lys Trp Lys Thr Leu Ala Arg Gln
425              430              435              440
GCC ATG GGA TAGGATGGAT CTTCACTCAA CAATCTCAAC GCCTTTCTTC 1393
Ala Met Gly
      443
GTCAAGTTGC ACACITTTTAA TCTGCTCAAA CAGCGGTTCA AATAAATATC CCCTTCCACT 1453
TAAAAAAAAA AAAAAAA 1470

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Sequence ID No.: 3

Sequence length: 2062

Sequence type: Nucleic acid

Number of strands: Double-strand

Topology: Straight chain

Source:

Biological name: Verbena (Verbena hybrida)

Tissue type: Petal

Direct source:

Library name: cDNA library

Clone name: pSHGT8

Sequence:

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ATTTTACCAA AAAAATAAAAA AAAAA ATG AGC AGA GCT CAC GTC CTC TTG GCC      52
                               Met Ser Arg Ala His Val Leu Leu Ala
                               1           5

ACA TTC CCA GCA CAG GGA CAC ATA AAT CCC GCC CTT CAA TTC GCC AAG      100
Thr Phe Pro Ala Gln Gly His Ile Asn Pro Ala Leu Gln Phe Ala Lys
 10           15           20           25

CGT CTC GCA AAT GCC GAC ATT CAA GTC ACA TTC TTC ACC AGC GTC TAC      148
Arg Leu Ala Asn Ala Asp Ile Gln Val Thr Phe Phe Thr Ser Val Tyr
           30           35           40

GCA TGG CGC CGC ATG TCC AGA ACC GCC GCT GGC TCA AAC GGG CTC ATC      196
Ala Trp Arg Arg Met Ser Arg Thr Ala Ala Gly Ser Asn Gly Leu Ile
           45           50           55

AAT TTT GTG TCG TTT TCC GAC GGG TAT GAC GAC GGG TTA CAG CCC GGA      244
Asn Phe Val Ser Phe Ser Asp Gly Tyr Asp Asp Gly Leu Gln Pro Gly
           60           65           70

GAC GAT GGG AAG AAC TAC ATG TCG GAG ATG AAA AGC AGA GGT ATA AAA      292
Asp Asp Gly Lys Asn Tyr Met Ser Glu Met Lys Ser Arg Gly Ile Lys
           75           80           85

GCC TTG AGC GAT ACT CTT GCA GCC AAT AAT GTC GAT CAA AAA AGC AGC      340
Ala Leu Ser Asp Thr Leu Ala Ala Asn Asn Val Asp Gln Lys Ser Ser
 90           95           100           105

AAA ATC ACG TTC GTG GTG TAC TCC CAC CTC TTT GCA TGG GCG GCC AAG      388
Lys Ile Thr Phe Val Val Tyr Ser His Leu Phe Ala Trp Ala Ala Lys
           110           115           120

GTG GCG CGT GAG TTC CAT CTC CGG AGC GCG CTA CTC TGG ATT GAG CCA      436
Val Ala Arg Glu Phe His Leu Arg Ser Ala Leu Leu Trp Ile Glu Pro
           125           130           135

GCT ACG GTG TTG GAT ATA TTT TAC TTT TAT TTC AAC GGC TAT AGC GAC      484
Ala Thr Val Leu Asp Ile Phe Tyr Phe Tyr Phe Asn Gly Tyr Ser Asp
           140           145           150

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GAA ATC GAT GCG GGT TCG GAT GCT ATT CAC TTG CCC GGA GGA CTC CCA	532
Glu Ile Asp Ala Gly Ser Asp Ala Ile His Leu Pro Gly Gly Leu Pro	
155 160 165	
GTG CTG GCC CAG CGT GAT TTA CCG TCT TTC CTT CTT CCT TCC ACG CAT	580
Val Leu Ala Gln Arg Asp Leu Pro Ser Phe Leu Leu Pro Ser Thr His	
170 175 180 185	
GAG AGA TTC CGT TCA CTG ATG AAG GAG AAA TTG GAA ACT TTA GAA GGT	628
Glu Arg Phe Arg Ser Leu Met Lys Glu Lys Leu Glu Thr Leu Glu Gly	
190 195 200	
GAA GAA AAA CCT AAG GTC TTG GTG AAC AGC TTT GAT GCG TTG GAG CCT	676
Glu Glu Lys Pro Lys Val Leu Val Asn Ser Phe Asp Ala Leu Glu Pro	
205 210 215	
GAT GCG CTC AAG GCC ATT GAT AAG TAC GAG ATG ATT GCA ATC GGG CCG	724
Asp Ala Leu Lys Ala Ile Asp Lys Tyr Glu Met Ile Ala Ile Gly Pro	
220 225 230	
TTG ATT CCT TCC GCA TTC TTG GAC GGT AAA GAT CCT TCG GAC AGG TCT	772
Leu Ile Pro Ser Ala Phe Leu Asp Gly Lys Asp Pro Ser Asp Arg Ser	
235 240 245	
TTC GGC GGA GAT TTG TTC GAG AAA GGG TCG AAT GAC GAC GAT TGC CTC	820
Phe Gly Gly Asp Leu Phe Glu Lys Gly Ser Asn Asp Asp Asp Cys Leu	
250 255 260 265	
GAA TGG TTG AGC ACG AAT CCT CGA TCT TCG GTG GTT TAC GTT TCG TTC	868
Glu Trp Leu Ser Thr Asn Pro Arg Ser Ser Val Val Tyr Val Ser Phe	
270 275 280	
GGA AGC TTC GTT AAT ACG ACG AAG TCG CAA ATG GAA GAG ATA GCA AGA	916
Gly Ser Phe Val Asn Thr Thr Lys Ser Gln Met Glu Glu Ile Ala Arg	
285 290 295	
GGG CTG TTA GAT TGT GGG AGG CCG TTT TTG TGG GTG GTA AGA GTA AAC	964
Gly Leu Leu Asp Cys Gly Arg Pro Phe Leu Trp Val Val Arg Val Asn	
300 305 310	
GAA GGA GAA GAG GTA TTG ATA AGT TGC ATG GAG GAG TTG AAA CGA GTG	1012
Glu Gly Glu Glu Val Leu Ile Ser Cys Met Glu Glu Leu Lys Arg Val	
315 320 325	
GGG AAA ATT GTA TCT TGG TGT TCT CAA TTG GAA GTC CTG ACG CAT CCC	1060
Gly Lys Ile Val Ser Trp Cys Ser Gln Leu Glu Val Leu Thr His Pro	
330 335 340 345	

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TCG TTG GGA TGT TTC GTG ACA CAC TGC GGG TGG AAT TCG ACT CTA GAG 1108
Ser Leu Gly Cys Phe Val Thr His Cys Gly Trp Asn Ser Thr Leu Glu
          350          355          360
AGT ATA TCT TTC GGG GTT CCG ATG GTG GCT TTT CCG CAG TGG TTC GAT 1156
Ser Ile Ser Phe Gly Val Pro Met Val Ala Phe Pro Gln Trp Phe Asp
          365          370          375
CAA GGG ACG AAT GCG AAG CTG ATG GAG GAT GTG TGG AGG ACG GGT GTG 1204
Gln Gly Thr Asn Ala Lys Leu Met Glu Asp Val Trp Arg Thr Gly Val
          380          385          390
AGA GTG AGA GCT AAT GAG GAG GGT AGC GTC GTT GAT GGT GAT GAA ATT 1252
Arg Val Arg Ala Asn Glu Glu Gly Ser Val Val Asp Gly Asp Glu Ile
          395          400          405
AGG AGA TGT ATT GAG GAG GTT ATG GAT GGG GGA GAA AAG AGT AGG AAA 1300
Arg Arg Cys Ile Glu Glu Val Met Asp Gly Gly Glu Lys Ser Arg Lys
          410          415          420          425
CTT AGA GAG AGT GCT GGC AAG TGG AAG GAT TTG GCA AGA AAA GCT ATG 1348
Leu Arg Glu Ser Ala Gly Lys Trp Lys Asp Leu Ala Arg Lys Ala Met
          430          435          440
GAG GAA GAT GGA TCT TCA GTT AAC AAC CTC AAG GTC TTT CTT GAT GAG 1396
Glu Glu Asp Gly Ser Ser Val Asn Asn Leu Lys Val Phe Leu Asp Glu
          445          450          455
GTT GTA GGT ATC TAAAGACGTA AATGAGGTCC CCATAGGCAA AATTGCAAAT 1448
Val Val Gly Ile
          460 461
TTCATCTCGT AAGTTGAATA CTTTTTGGCT TTAATTTTGT TCGAGTTTGT TTTTCAAAAT 1508
TTATCTTGTA ATTTTACATT GAGTGTAAT TTAGTCTGAT TTAACTGGA AAAATATAAA 1568
ATTCATTGTT GAGACTCTTC ATCAAAATCA TCTGATTTCC TTTATTGTCT TGGTCAAAAT 1628
TCTCATATCA ATTGGAAAAA ATAAATTTCA AAATCGTCCA ATTTTGAACC AAGAAAGAAG 1688
TATAATTTGA CAAAATAAT AAAAGGATTC AAGTGATCTT GATGAAGTGT CTGAGCGACG 1748
AGTTCTATAT TTTTCCACCG AATTTCTAAC GAGTTTTTGA ATTTTTTTTA GCCAAAATCG 1808
GACTAACTTT GTACAAAATG AAAAGTTATA TGATGAAATT TAAAAAACA AACTCAGACA 1868
ATAATAAAGC CCGAAAGTAG TAAAATTACC TGACGAAATT TGCAATTTTC CCTCCTATTT 1928
TAATTTTTTT GGTGTGTTTA ATAAATCGGT TATTTTACTT TTAATTAAAA TAAAAGTGAG 1988
ATGCATGATA GCTTGGTGAG TATATATGAG TTGATGGTAA TGTACGATAT TTTCTAAAAA 2048
AAAAAAAAAA AAAA 2062

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Sequence ID No.: 4

Sequence length: 1671

Sequence type: Nucleic acid  
Number of strands: Double-strand  
Topology: Straight chain  
Source:

Biological name: Trenia

Tissue type: Petal

Direct source:

Library name: cDNA library

Clone name: pSTGT5

Sequence:

AACACATAAA	AAAAAAATAA	AAGAAGAAAT	AATTAAAAAA	AAAA	ATG GTT AAC	53
					Met Val Asn	
					1	
AAA CGC CAT ATT CTA CTA GCA ACA TTC CCA GCA CAA GGC CAC ATA AAC						101
Lys Arg His Ile Leu Leu Ala Thr Phe Pro Ala Gln Gly His Ile Asn						
5	10			15		
CCT TCT CTC GAG TTC GCC AAA AGG CTC CTC AAC ACC GGA TAC GTC GAC						149
Pro Ser Leu Glu Phe Ala Lys Arg Leu Leu Asn Thr Gly Tyr Val Asp						
20	25		30		35	
CAA GTC ACA TTC TTC ACG AGT GTA TAC GCA TTG AGA CGC ATG CGC TTC						197
Gln Val Thr Phe Phe Thr Ser Val Tyr Ala Leu Arg Arg Met Arg Phe						
	40		45		50	
GAA ACC GAT CCG AGC AGC AGA ATC GAT TTC GTG GCA TKT YCA GAT TCT						245
Glu Thr Asp Pro Ser Ser Arg Ile Asp Phe Val Ala Xaa Xaa Asp Ser						
	55		60		65	
TAC GAT GAT GGC TTA AAG AAA GGC GAC GAT GGC AAA AAC TAC ATG TCG						293
Tyr Asp Asp Gly Leu Lys Lys Gly Asp Asp Gly Lys Asn Tyr Met Ser						
	70		75		80	
GAG ATG AGA AAG CGC GGA ACG AAG GCC TTA AAG GAC ACT CTT ATT AAG						341
Glu Met Arg Lys Arg Gly Thr Lys Ala Leu Lys Asp Thr Leu Ile Lys						
	85		90		95	
CTC AAC GAT GCT GCG ATG GGA AGT GAA TGT TAC AAT CGC GTG AGC TTT						389
Leu Asn Asp Ala Ala Met Gly Ser Glu Cys Tyr Asn Arg Val Ser Phe						
100		105		110		115

GTG GTG TAC TCT CAT CTA TTT TCG TGG GCA GCT GAA GTG GCG CGT GAA	437
Val Val Tyr Ser His Leu Phe Ser Trp Ala Ala Glu Val Ala Arg Glu	
120 125 130	
GTC GAC GTG CCG AGT GCC CTT CTT TGG ATT GAA CCG GCT ACG GTT TTC	485
Val Asp Val Pro Ser Ala Leu Leu Trp Ile Glu Pro Ala Thr Val Phe	
135 140 145	
GAT GTG TAC TAT TTT TAC TTC AAT GGG TAT GCC GAT GAT ATC GAT GCG	533
Asp Val Tyr Tyr Phe Tyr Phe Asn Gly Tyr Ala Asp Asp Ile Asp Ala	
150 155 160	
GGC TCA GAT CAA ATC CAA CTG CCC AAT CTT CCG CAG CTC TCC AAG CAA	581
Gly Ser Asp Gln Ile Gln Leu Pro Asn Leu Pro Gln Leu Ser Lys Gln	
165 170 175	
GAT CTC CCC TCT TTC CTA CTC CCT TCG AGC CCC GCG AGA TTC CGA ACC	629
Asp Leu Pro Ser Phe Leu Leu Pro Ser Ser Pro Ala Arg Phe Arg Thr	
180 185 190 195	
CTA ATG AAA GAA AAG TTC GAC ACG CTC GAC AAA GAA CCG AAA GCG AAG	677
Leu Met Lys Glu Lys Phe Asp Thr Leu Asp Lys Glu Pro Lys Ala Lys	
200 205 210	
GTC TTG ATA AAC ACG TTC GAC GCA TTA GAA ACC GAA CAA CTC AAA GCC	725
Val Leu Ile Asn Thr Phe Asp Ala Leu Glu Thr Glu Gln Leu Lys Ala	
215 220 225	
ATC GAC AGG TAT GAA CTA ATA TCC ATC GGC CCA TTA ATC CCA TCA TCG	773
Ile Asp Arg Tyr Glu Leu Ile Ser Ile Gly Pro Leu Ile Pro Ser Ser	
230 235 240	
ATA TTC TCA GAT GGC AAC GAC CCC TCA TCA AGC AAC AAA TCC TAC GGT	821
Ile Phe Ser Asp Gly Asn Asp Pro Ser Ser Ser Asn Lys Ser Tyr Gly	
245 250 255	
GGA GAC CTC TTC AGA AAA GCC GAT GAA ACT TAC ATG GAC TGG CTA AAC	869
Gly Asp Leu Phe Arg Lys Ala Asp Glu Thr Tyr Met Asp Trp Leu Asn	
260 265 270 275	
TCA AAA CCC GAA TCA TCG GTC GTT TAC GTT TCG TTC GGG AGC CTC CTG	917
Ser Lys Pro Glu Ser Ser Val Val Tyr Val Ser Phe Gly Ser Leu Leu	
280 285 290	
AGG CTC CCG AAA CCC CAA ATG GAA GAA ATA GCA ATA GGG CTT TCA GAC	965
Arg Leu Pro Lys Pro Gln Met Glu Glu Ile Ala Ile Gly Leu Ser Asp	
295 300 305	

[illegible]

Sequence ID No.: 5  
Sequence length: 1437  
Sequence type: Nucleic acid  
Number of strands: Double-strand  
Topology: Straight chain  
Source:

Biological name: Perilla (*Perilla frutescens*)  
Tissue type: Leaf

Direct source:

Library name: cDNA library  
Clone name: p8R6

Sequence:

TTCAAAACTC	ATAACGTGAT	TGAGCTAATG	TGCACATCTT	CCTCTTCAAA	GTCTACAGTG	60
TCATCCTACC	AGCATCATCA	TGATCAATCT	CTTTATAATG	AGGAGAATGG	AGTAACAAGG	120
AGTGGGTTTT	GTTACTCAGC	TTCAACCTAC	GTACGTACTA	CTACTGACTC	AACTCTCAAG	180
AGAATGAATA	TAATATATAA	TGGGCGATAG	ATCTTTGTAG	ATATGTAGGT	GTAGCCTGCA	240
GGTGGTTAAT	TAATTTCCGG	TGTGGGAAAA	TAAATAAATA	AATAAATATA	GCG ATG AGC	299
					Met Ser	
					1	
AGC AGC AGC AGC	AGA AGG TGG AGA	GAG AAT GAG GGG	ATG CGA AGG ACA			347
Ser Ser Ser Ser	Arg Arg Trp Arg	Glu Asn Glu Gly	Met Arg Arg Thr			
5	10	15				
TTG CTG GGG TTG	GGT TTG GGG CAG	TTG GTT TCT TTC	GAT TTG GCT ATC			395
Leu Leu Gly Leu	Gly Leu Gly Gln	Leu Val Ser Phe	Asp Leu Ala Ile			
20	25	30				
ATG ACC TTT TCT	GCT TCT TTG GTT	TCA ACC ACA GTG	GAT GCA CCA CTT			443
Met Thr Phe Ser	Ala Ser Leu Val	Ser Thr Thr Val	Asp Ala Pro Leu			
35	40	45	50			
ACT ATG TCG TTC	ACT ACA TAC ACT	GTT GTG GCC CTG	CTC TAT GGA ACC			491
Thr Met Ser Phe	Thr Thr Tyr Thr	Val Val Ala Leu	Leu Tyr Gly Thr			
	55	60	65			
ATC TTG CTT TAC	CGC CGC CAC AAA	TTC TTG GTT CCA	TGG TAC TGG TAT			539
Ile Leu Leu Tyr	Arg Arg His Lys	Phe Leu Val Pro	Trp Tyr Trp Tyr			
70	75	80				

GCT	CTC	CTG	GGG	TTC	GTG	GAC	GTC	CAC	GGC	AAT	TAT	CTT	GTT	AAT	AAA	587
Ala	Leu	Leu	Gly	Phe	Val	Asp	Val	His	Gly	Asn	Tyr	Leu	Val	Asn	Lys	
85						90						95				
GCA	TTC	GAG	TTG	ACA	TCG	ATT	ACG	AGT	GTG	AGC	ATA	CTG	GAT	TGT	TGG	635
Ala	Phe	Glu	Leu	Thr	Ser	Ile	Thr	Ser	Val	Ser	Ile	Leu	Asp	Cys	Trp	
100						105						110				
ACA	ATC	GTG	TGG	TCC	ATC	ATC	TTT	ACA	TGG	ATG	TTC	CTA	GGC	ACA	AAA	683
Thr	Ile	Val	Trp	Ser	Ile	Ile	Phe	Thr	Trp	Met	Phe	Leu	Gly	Thr	Lys	
115						120						125			130	
TAC	TCT	GTA	TAC	CAG	TTT	GTC	GGT	GCT	GCT	ATT	TGT	GTA	GGA	GGC	CTC	731
Tyr	Ser	Val	Tyr	Gln	Phe	Val	Gly	Ala	Ala	Ile	Cys	Val	Gly	Gly	Leu	
			135						140						145	
CTC	CTC	GTG	CTT	CTT	TCC	GAC	TCA	GGG	GTC	ACT	GCT	GCT	GGT	TCG	AAT	779
Leu	Leu	Val	Leu	Leu	Ser	Asp	Ser	Gly	Val	Thr	Ala	Ala	Gly	Ser	Asn	
			150						155						160	
CCT	CTT	TTG	GGT	GAT	TTT	CTT	GTC	ATA	ACA	GGC	TCT	ATT	TTG	TTC	ACA	827
Pro	Leu	Leu	Gly	Asp	Phe	Leu	Val	Ile	Thr	Gly	Ser	Ile	Leu	Phe	Thr	
165						170						175				
CTC	AGC	ACT	GTT	GGT	CAG	GAA	TAC	TGC	GTG	AAG	AGG	AAA	GAT	CGT	ATT	875
Leu	Ser	Thr	Val	Gly	Gln	Glu	Tyr	Cys	Val	Lys	Arg	Lys	Asp	Arg	Ile	
180						185						190				
GAA	GTA	GTA	GCA	ATG	ATC	GGT	GTA	TTT	GGT	ATG	CTC	ATC	AGT	GCA	ACC	923
Glu	Val	Val	Ala	Met	Ile	Gly	Val	Phe	Gly	Met	Leu	Ile	Ser	Ala	Thr	
195						200						205			210	
GAG	ATT	ACT	GTG	CTG	GAG	AGG	AAT	GCC	CTC	TCA	TCA	ATG	CAG	TGG	TCT	971
Glu	Ile	Thr	Val	Leu	Glu	Arg	Asn	Ala	Leu	Ser	Ser	Met	Gln	Trp	Ser	
			215						220						225	
ACT	GGA	CTT	TTG	GCA	GCC	TAT	GTT	GTT	TAT	GCA	CTG	TCC	AGC	TTC	CTC	1019
Thr	Gly	Leu	Leu	Ala	Ala	Tyr	Val	Val	Tyr	Ala	Leu	Ser	Ser	Phe	Leu	
			230						235						240	
TTC	TGC	ACA	CTC	ACC	CCT	TTT	CTT	CTC	AAG	ATG	AGT	GGC	GCT	GCA	TTT	1067
Phe	Cys	Thr	Leu	Thr	Pro	Phe	Leu	Leu	Lys	Met	Ser	Gly	Ala	Ala	Phe	
245						250						255				
TTC	AAT	CTT	TCC	ATG	CTT	ACA	TCT	GAT	ATG	TGG	GCT	GTT	GCA	ATT	AGG	1115
Phe	Asn	Leu	Ser	Met	Leu	Thr	Ser	Asp	Met	Trp	Ala	Val	Ala	Ile	Arg	
260						265						270				

ACA TTC ATA TAC AAC CAG GAG GTT GAT TGG TTA TAC TAT TTG GCC TTT 1163  
Thr Phe Ile Tyr Asn Gln Glu Val Asp Trp Leu Tyr Tyr Leu Ala Phe  
275 280 285 290  
TGT CTC GTT GTT GTT GGA ATA TTC ATA TAT ACA AAA ACA GAG AAG GAT 1211  
Cys Leu Val Val Val Gly Ile Phe Ile Tyr Thr Lys Thr Glu Lys Asp  
295 300 305  
CCT AAC AAT ACG AGA GCC CTT GAG AAT GGA AAC TTG GAT CAT GAA TAT 1259  
Pro Asn Asn Thr Arg Ala Leu Glu Asn Gly Asn Leu Asp His Glu Tyr  
310 315 320  
AGT CTC CTT GAG GAT CAA GAT GAC ACA CCA AGA AAA CCA TAGCTAGCTT 1308  
Ser Leu Leu Glu Asp Gln Asp Asp Thr Pro Arg Lys Pro  
325 330 335  
TGCCCAACAAT CTTTTCATCA ACAGTTTAA ATAATTCGTG AGGGGGAGAG AGATCGAGAT 1368  
ACTAATTAAT GGACGTCTAT TATATAGTTG GAGGTTTTTG TTTATTTAT TTATTTGAGT 1428  
AAAAAAAAA 1437

Sequence ID No.: 6

Sequence length: 2105

Sequence type: Nucleic acid

Number of strands: Double-strand

Topology: Straight chain

Source:

Biological name: Petunia

Tissue type: Leaf

Direct source:

Library name: cDNA library

Clone name: pSPGT1

Sequence:

AGTGAGCGCA ACGCAATTAA TGTGAGTTAG CTCACTCATT AGGCACCCCA GGCTTTACAC 60  
TTTATGCTTC CGGCTCGTAT GTTGTGTGGA ATTGTGAGCG GATAACAATT TCACACAGGA 120  
AACAGCTATG ACCATGATTA CGCCAAGCTC GAAATTAACC CTCACTAAAG GGAACAAAAG 180  
CTGGAGCTCC ACGCGGTGGC GGCCGCTCTA GAACTAGTGG ATCCCCCGGG CTGCAGGAAT 240  
TCCGTTGCTG TCGCCACAAT TTACAAACCA AGAAATTAAG CATCCCTTTC CCCCCCTTAA 300  
AAAACATACA AGTTTTTAAT TTTTCACTAA GCAAGAAAAT ATG GTG CAG CCT CAT GTC 358

Met Val Gln Pro His Val



ATC TTA ACA ACA TTT CCA GCA CAA GGC CAT ATT AAT CCA GCA CTT CAA	406
Ile Leu Thr Thr Phe Pro Ala Gln Gly His Ile Asn Pro Ala Leu Gln	
10 15 20	
TTT GCC AAG AAT CTT GTC AAG ATG GGC ATA GAA GTG ACA TTT TCT ACA	454
Phe Ala Lys Asn Leu Val Lys Met Gly Ile Glu Val Thr Phe Ser Thr	
25 30 35	
AGC ATT TAT GCC CAA AGC CGT ATG GAT GAA AAA TCC ATT CTT AAT GCA	502
Ser Ile Tyr Ala Gln Ser Arg Met Asp Glu Lys Ser Ile Leu Asn Ala	
40 45 50	
CCA AAA GGA TTG AAT TTC ATT CCA TTT TCC GAT GGC TTT GAT GAA GGT	550
Pro Lys Gly Leu Asn Phe Ile Pro Phe Ser Asp Gly Phe Asp Glu Gly	
55 60 65 70	
TTT GAT CAT TCA AAA GAC CCT GTA TTT TAC ATG TCA CAA CTT CGT AAA	598
Phe Asp His Ser Lys Asp Pro Val Phe Tyr Met Ser Gln Leu Arg Lys	
75 80 85	
TGT GGA AGT GAA ACT GTC AAA AAA ATA ATT CTC ACT TGC TCT GAA AAT	646
Cys Gly Ser Glu Thr Val Lys Lys Ile Ile Leu Thr Cys Ser Glu Asn	
90 95 100	
GGA CAG CCT ATA ACT TGC CTA CTT TAC TCC ATT TTC CTT CCT TGG GCA	694
Gly Gln Pro Ile Thr Cys Leu Leu Tyr Ser Ile Phe Leu Pro Trp Ala	
105 110 115	
GCA GAG GTA GCA CGT GAA GTT CAC ATC CCT TCT GCT CTT CTT TGG AGT	742
Ala Glu Val Ala Arg Glu Val His Ile Pro Ser Ala Leu Leu Trp Ser	
120 125 130	
CAA CCA GCA ACA ATA TTG GAC ATA TAT TAC TTC AAC TTT CAT GGA TAT	790
Gln Pro Ala Thr Ile Leu Asp Ile Tyr Tyr Phe Asn Phe His Gly Tyr	
135 140 145 150	
GAA AAA GCT ATG GCT AAT GAA TCC AAT GAT CCA AAT TGG TCC ATT CAA	838
Glu Lys Ala Met Ala Asn Glu Ser Asn Asp Pro Asn Trp Ser Ile Gln	
155 160 165	
CTT CCC GGG CTT CCA CTA CTG GAA ACT CGA GAT CTT CCT TCA TTT TTA	886
Leu Pro Gly Leu Pro Leu Leu Glu Thr Arg Asp Leu Pro Ser Phe Leu	
170 175 180	
CTT CCT TAT GGT GCA AAA GGG AGT CTT CGA GTT GCA CTT CCA CCA TTC	934
Leu Pro Tyr Gly Ala Lys Gly Ser Leu Arg Val Ala Leu Pro Pro Phe	
185 190 195	

15442229" 5562450

AAA GAA TTG ATA GAC ACA TTA GAT GCT GAA ACC ACT CCT AAG ATT CTT	982
Lys Glu Leu Ile Asp Thr Leu Asp Ala Glu Thr Thr Pro Lys Ile Leu	
200 205 210	
GTG AAT ACA TTT GAT GAA TTA GAG CCT GAG GCA CTC AAT GCA ATT GAA	1030
Val Asn Thr Phe Asp Glu Leu Glu Pro Glu Ala Leu Asn Ala Ile Glu	
215 220 225 230	
GGT TAT AAG TTT TAT GGA ATT GGA CCG TTG ATT CCT TCT GCT TTC TTG	1078
Gly Tyr Lys Phe Tyr Gly Ile Gly Pro Leu Ile Pro Ser Ala Phe Leu	
235 240 245	
GGT GGA AAT GAC CCT TTA GAT GCT TCA TTT GGT GGT GAT CTT TTT CAA	1126
Gly Gly Asn Asp Pro Leu Asp Ala Ser Phe Gly Gly Asp Leu Phe Gln	
250 255 260	
AAT TCA AAT GAC TAT ATG GAA TGG TTA AAC TCA AAG CCA AAT TCA TCA	1174
Asn Ser Asn Asp Tyr Met Glu Trp Leu Asn Ser Lys Pro Asn Ser Ser	
265 270 275	
GTT GTT TAT ATA TCT TTT GGG AGT CTA ATG AAT CCA TCT ATT AGC CAA	1222
Val Val Tyr Ile Ser Phe Gly Ser Leu Met Asn Pro Ser Ile Ser Gln	
280 285 290	
ATG GAG GAG ATA TCA AAA GGG TTG ATA GAC ATA GGA AGG CCG TTT TTA	1270
Met Glu Glu Ile Ser Lys Gly Leu Ile Asp Ile Gly Arg Pro Phe Leu	
295 300 305 310	
TGG GTG ATA AAA GAA AAT GAA AAA GGC AAA GAA GAA GAG AAT AAA AAG	1318
Trp Val Ile Lys Glu Asn Glu Lys Gly Lys Glu Glu Glu Asn Lys Lys	
315 320 325	
CTT GGT TGT ATT GAA GAA TTG GAA AAA ATA GGA AAA ATA GTT CCA TGG	1366
Leu Gly Cys Ile Glu Glu Leu Glu Lys Ile Gly Lys Ile Val Pro Trp	
330 335 340	
TGT TCA CAA CTT GAA GTT CTA AAA CAT CCA TCT TTA GGA TGT TTT GTT	1414
Cys Ser Gln Leu Glu Val Leu Lys His Pro Ser Leu Gly Cys Phe Val	
345 350 355	
TCT CAT TGT GGA TGG AAT TCA GCC TTA GAG AGT TTA GCT TGT GGA GTG	1462
Ser His Cys Gly Trp Asn Ser Ala Leu Glu Ser Leu Ala Cys Gly Val	
360 365 370	
CCA GTT GTG GCA TTT CCT CAA TGG ACA GAT CAA ATG ACA AAT GCC AAA	1510
Pro Val Val Ala Phe Pro Gln Trp Thr Asp Gln Met Thr Asn Ala Lys	
375 380 385 390	

CCNY "BIOCHEM"

Table 1. Demographic characteristics of the study population	
Age (years)	65.5 ± 1.2
Gender (male/female)	10/10
Education (years)	12.5 ± 0.5
Occupation (white/blue)	10/10
Marital status (married/divorced/widowed)	10/10/0
Smoking status (smoker/nonsmoker)	10/10
Alcohol consumption (yes/no)	10/10
Comorbidities (hypertension/diabetes/cholesterol)	10/10/10
Medication (antihypertensive/antidiabetic/anticholesterol)	10/10/10
Family history (hypertension/diabetes/cholesterol)	10/10/10
Physical activity (yes/no)	10/10
Stress level (high/low)	10/10
Social support (yes/no)	10/10
Quality of life (yes/no)	10/10
Health status (good/poor)	10/10
Life expectancy (years)	15.5 ± 0.5
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Healthcare quality (yes/no)	10/10
Healthcare satisfaction (yes/no)	10/10
Healthcare utilization (yes/no)	10/10
Health insurance (yes/no)	10/10
Healthcare costs (yes/no)	10/10
Healthcare access (yes/no)	10/10
Health	

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## Declaration and Power of Attorney For Patent Application

### 特許出願宣言書及び委任状

### Japanese Language Declaration

### 日本語宣言書

下記の氏名の発明者として、私は以下の通り宣言します。

As a below named inventor, I hereby declare that:

私の住所、私書箱、国籍は下記の私の氏名の後に記載された通りです。

My residence, post office address and citizenship are as stated next to my name.

下記の名称の発明に関して請求範囲に記載され、特許出願している発明内容について、私が最初かつ唯一の発明者（下記の氏名が一つの場合）もしくは最初かつ共同発明者であると（下記の名称が複数の場合）信じています。

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled

GENE CODING FOR A PROTEIN HAVING

GLYCOSIDE TRANSFER ACTIVITY

上記発明の明細書（下記の欄でx印がついていない場合は、本書に添付）は、

the specification of which is attached hereto unless the following box is checked:

☐ 月 日に提出され、米国出願番号または特許協定条約国際出願番号を \_\_\_\_\_ とし、  
（該当する場合） \_\_\_\_\_ に訂正されました。

☒ was filed on July 16, 1998  
as United States Application Number or  
PCT International Application Number  
PCT/JP98/03199 and was amended on \_\_\_\_\_  
(if applicable).

私は、特許請求範囲を含む上記訂正後の明細書を検討し、内容を理解していることをここに表明します。

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by any amendment referred to above.

私は、連邦規則法典第37編第1条56項に定義されたとおり、特許資格の有無について重要な情報を開示する義務があることを認めます。

I acknowledge the duty to disclose information which is material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

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## Japanese Language Declaration (日本語宣言書)

私は、米国法典第35編119条(a)-(d)項又は365条(b)項に基づき下記の、米国外の国の少なくとも一カ国を指定している特許協力条約365(a)項に基づく国際出願、又は外国での特許出願もしくは発明者証の出願についての外国優先権をここに主張するとともに、優先権を主張している、本出願の前に出願された特許または発明者証の外国出願を以下に、枠内をマークすることで、示しています。

### Prior Foreign Application(s)

外国での先行出願 9-200571 (Pat. Appln.) (Number) (番号)	Japan (Country) (国名)
(Number) (番号)	(Country) (国名)

私は、第35編米国法典119条(e)項に基づいて下記の米国外の特許出願規定に記載された権利をここに主張いたします。

(Application No.) (出願番号)	(Filing Date) (出願日)
-----------------------------	------------------------

私は、下記の米国法典第35編120条に基づいて下記の米国外の特許出願に記載された権利、又は米国外を指定している特許協力条約365条(c)に基づき権利をここに主張します。また、本出願の各請求範囲の内容が米国法典第35編112条第1項又は特許協力条約で規定された方法で先行する米国外の特許出願に開示されていない限り、その先行米国外出願書提出日以降で本出願書の日本国内または特許協力条約国際提出日までの期間中に入手された、連邦規則法典第37編1条56項で定義された特許資格の有無に関する重要な情報について開示義務があることを認識しています。

(Application No.) (出願番号)	(Filing Date) (出願日)
-----------------------------	------------------------

(Application No.) (出願番号)	(Filing Date) (出願日)
-----------------------------	------------------------

私は、私自身の知識に基づいて本宣言書中で私が行なう表明が真実であり、かつ私の入手した情報と私の信じることに基づき表明が全て真実であると信じていること、さらに故意になされた虚偽の表明及びそれと同等の行為は米国法典第18編第1001条に基づき、罰金または拘禁、もしくはその両方により処罰されること、そしてそのような故意による虚偽の表明を行えば、出願した、又は既に許可された特許の有効性が失われることを認識し、よってここに上記のごとく宣誓を致します。

I hereby claim foreign priority under Title 35, United States Code, Section 119 (a)-(d) or 365(b) of any foreign application(s) for patent or inventor's certificate, or 365(a) of any PCT International application which designated at least one country other than the United States, listed below and have also identified below, by checking the box, any foreign application for patent or inventor's certificate, or PCT International application having a filing date before that of the application on which priority is claimed.

### Priority Not Claimed

優先権主張なし

25/July/1997  
(Day/Month/Year Filed)  
(出願年月日)

(Day/Month/Year Filed)  
(出願年月日)

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below.

(Application No.) (出願番号)	(Filing Date) (出願日)
-----------------------------	------------------------

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States application(s), or 365(c) of any PCT International application designating the United States, listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States or PCT International application in the manner provided by the first paragraph of Title 35, United States Code Section 112, I acknowledge the duty to disclose information which is material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56 which became available between the filing date of the prior application and the national or PCT International filing date of application.

(Status: Patented, Pending, Abandoned) (現況: 特許許可済、係属中、放棄済)
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(Status: Patented, Pending, Abandoned) (現況: 特許許可済、係属中、放棄済)
---

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

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## Japanese Language Declaration

(日本語宣言書)

委任状： 私は下記の発明者として、本出願に関する一切の  
手続を米特許商標局に対して遂行する弁理士または代理人  
として、下記の者を指名いたします。(弁理士、または代理  
人の氏名及び登録番号を明記のこと)

POWER OF ATTORNEY: As a named inventor, I hereby appoint  
the following attorney(s) and/or agent(s) to prosecute this  
application and transact all business in the Patent and Trademark  
Office connected therewith (list name and registration number)

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発明者の署名	日付	Inventor's signature	Date
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		Japanese	
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(第三以降の共同発明者についても同様に記載し、署名をす  
ること)

(Supply similar information and signature for third and subsequent  
joint inventors.)

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JPX

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第三共同発明者 日付	Third inventor's signature <u>[Signature]</u> Date March 12, 1999
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第四共同発明者 日付	Fourth inventor's signature <u>[Signature]</u> Date March 12, 1999
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JPX

400  
JPX

500  
JPX

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第六共同発明者 日付	Sixth inventor's signature <u>[Signature]</u> Date March 12, 1999
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(第七以降の共同発明者についても同様に  
記載し、署名をすること)

(Supply similar information and signature for  
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